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Pia Høybye-Olsen

PATENT- OG VAREMÆRKESTYRELSEN

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25 MRS. 2003
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# Methodologies for improving the quality of meat, health status of animals and impact on environment

All patent and non-patent references cited in the present application are hereby incorporated by reference in their entirety.

## Field of invention

The present invention relates to methods and compositions for improving the quality of meat, to methods and compositions for preventing or reducing male animal taint, primarily boar taint caused by skatole and/or androstenone. The invention also relates to methods for improving the health status of animals and to methods for reducing animal caused odours in general.

## Background of invention

### Boar taint

Boar taint is a large problem in agriculture. The phenomenon referred to as "boar taint" is an ill-defined complex problem from a causal mechanistic standpoint that is characterised in pork meat by off-odours and flavours from a human sensory perspective. In addition, the live animals that lead to boar taint in meat also impart highly unacceptable off-odours to their environmental surroundings.

Although the term "boar taint" implies that the problem is restricted to boars (sexually mature male pigs), the problem is by no means exclusive to such animals. Male pigs in general and to a lesser extent female and castrated male pigs also exhibit the phenomena associated with boar taint or pig off odour. In addition, the negative effects of bore taint increase with the increasing age of the animals.

Boar taint is generally believed to be caused by at least two contributing factors, skatole and androstenone (Bonneau et al., 2000; Dijksterhuis et al., 2000). Skatole is formed by microbial breakdown of tryptophane in the gastrointestinal tract of pigs, in particular in the colon and in the caecum. Androstenone is synthesised in the testicles. Both compounds are metabolised in the liver. Some boars have a lower

rate of metabolism in the liver and consequently these animals result in meat that contains boar taint to a higher extent than the average pig.

The phenomena associated with boar taint and/or pig off-odour is several. First and foremost the odour and flavour of pork meat is affected negatively in particular due to the presence of skatole and/or androstenone over certain levels. The odour and flavour may be affected to such an extent that the meat is not acceptable for human consumption. In addition, live (fattening) pigs are associated with an unpleasant odour caused by volatile microbial metabolites in their excreta. The unpleasant odours mainly stem from microbial produced volatiles in liquid manure (mixture of faeces and urine). Two important volatile components of liquid manure are p-Cresol and skatole plus ammonia. The net result of this aspect of the pig off-odour phenomena constitutes an environmental problem in terms of publicly unacceptable negative odours imparted to the surroundings of large pig farms (Hartung & Rokicki, 1984; Hidaka et al., 1986; Sutton et al., 1999). Ammonium evaporates as NH3 and acidifies the environment. Fixing of nitrogen to less volatile compounds during passage through the gastrointestinal tract would thus be desirable.

Danish (and other) slaughterhouses have set thresholds for the allowable amount of skatole in entire male pigs backfat. The limit today is 0.25 ppm of skatole in the backfat of entire male pigs. In the past, the limit was set to 0.20 ppm and with this limit approximately 8 % of all male pigs had to be discarded. With the present level of 0.25 ppm approximately 5 % of all male pigs are discarded as boar tainted meat. The meat is then used in sausage manufacture in association with boar taint free meat, such that the negative boar taint odours and flavours are masked and thus are not a problem in human acceptability terms any longer especially when eaten cold. However, in this context the pork meat does not realise its original potential economic value. Pigs with elevated skatole contents thus constitute a substantial economic loss to agriculture. Therefore, there is a large monetary incentive to reduce and minimise the percentage of animals with high levels of skatole in the pig population.

The limit of 0.20-0.25 ppm skatole has been more or less arbitrarily set thus far and in practice skatole also negatively affects the sensory properties of pork meat from entire male pigs at concentrations of as low as 0.15 ppm and maybe has negative

effects at even lower levels when in combination with higher concentrations of androstenone and other negative odorous compounds. Reducing the concentration of skatole below 0.15 ppm to as close to zero as possible will result in elevated quality of all pork meat from a human sensory perspective and consequently allow higher prices to be obtained for pork meat per se. In addition, the on-farm pig odour problems will also be reduced substantially with great benefit to the public.

**Existing methods for boar taint control**

Methods for boar taint control comprise castration of male pigs and feeding with inulin and fructooligosaccharides.

**Castration of male pigs**

The phenomena of boar taint associated negative effects has been addressed in the state of the art. The most common preventive measure is to castrate male pigs either physically through removal of the testicles during the first week of the male pig's life, or chemically through immunovaccination (Bonneau and Carelli, 1987). Immunological castration of male pigs with a synthetic aqueous vaccine is possible (Dunshea Et al., 2001). Immunization of pigs against gonadotrophin releasing factor (GnRF) prevents boar taint and affects boar growth and behaviour (Metz et al., 2002). Overall, today the costs of immunovaccination are prohibitively high. Furthermore, it is only allowed by authorities in a few countries (USA, Australia) due to welfare problems, and thus not a realistic alternative in other countries.

Physical castration is commonly carried out by the farmer without sedation or anaesthetics. The consequences of this include in some cases infections of the wounds with resulting costs for treating the higher level of infections in the stock. Moreover, physical castration is carried out rapidly and the efficiency is not always 100 %.

Castration reduces the boar taint problems of skatole and androstenone in the meat and fat, but it does not eliminate the negative effects. Furthermore, castration does not address the problem of elevated p-Cresol and skatole levels and live pig off-odour problems (stable and manure offensive-odour) found in all pig stables with especially fattening pigs.

It is expected that mass castration of piglets will be forbidden in the near future for reasons of animal welfare at least in the EU area. In Norway such castration is forbidden from 2009. In the interim period, authorised veterinarians can only perform castration. Castration by veterinarians makes the costs prohibitively high for industrial scale pig farming.

**Inulin and fructooligosaccharides (FOS)**

It is known that the production of skatole from tryptophan in the intestine can be reduced by feeding pigs with inulin (Claus, 1992; Claus 1994) and fructooligosaccharides, FOS, (see e.g. Jensen & Jensen, 1998; Knarreborg et al., 2002; Xu et al, 2002). However, to date a sufficient efficiency in reducing boar-taint remains to be demonstrated for these compounds. In for example Claus (1992), DE 42 23 051 it was demonstrated that the skatole content of backfat could be reduced only by 55% by feeding 140 kg pigs 2x35 g of inulin (from Dahlia tubers) daily.

**Live pig odour reduction**

The malodorous volatile compounds emitted from pig production units are an increasing problem in areas with intensive animal production. Several strategies for reduction of emission of odour have been tried e.g. (I) Biofilters, (II) Continuous aerobic treatment, (III) using oil and foam layers, (IV) additives to manure (e.g. acids), and (V) feed or change of feed composition. Although some improvement in ambient air quality has been obtained by these methods, none of them have found widespread use in practical conditions.

The solution for odour reduction should both be economically feasible and fit into the production systems without major investments. In addition the quality of the resulting meat product should remain at the same level, ideally with an increased product quality.

The most efficient solution would be to stop the production of malodorous compounds before the compounds end up in the manure, i.e. in the pig itself. This should be achieved with a suitable feed composition, which changes the spectrum of produced odorous compounds so the odour impression is changed to a less disagreeable composition. The need for investment in mechanical deodorising equipment in connection with the stable can therefore be omitted.

The odour active compounds originate from microbial degradation of residual feed components in the manure. The odour compounds can be divided in two groups depending on their origin: (I) compounds from fermentation of carbohydrates, and (II) compounds originating from fermentation of proteins. Degradation compounds from fermentable carbohydrates are usually short chain fatty acids (acetic acid, propionic acid, butyric acid and valeric acid) and short chain alcohols. The degradation products from proteins are a more complex mixture. They are branched short chain fatty acids (isobutyric acid), indoles (skatole and indole), phenols (p-cresol) and sulfur compounds (hydrogen sulfide, dimethyl disulfide). The compounds from the last group (protein fermentation products) have more disagreeable odours than the first group (carbohydrate fermentation products) and lower odour thresholds. This means they have a relatively higher negative impact on the air quality. The compounds produced can also be combined with each other e.g. volatile fatty acids can be combined with alcohols and result in esters which have other odour characteristics usually with less offensive odour notes. This process is facilitated by esterases, which can be produced by microorganisms.

The strategy for changing the composition of the odour active compounds (and thereby increase the air quality) would then be to increase the amount of less odour offensive compounds (from carbohydrate degradation) at the expense of the more odour active compounds (from protein degradation). If the odour active compounds also include synthesis of esters the odour quality would be further improved.

Accordingly there is need in the art for developing methods which are compatible with modern industrial scale farming for addressing the problems of taint in animals especially taint in male animals, primarily boar taint, including stable malodour, and meat taste.

**Control of parasite infections in pigs the state of the art**

Infections with intestinal parasites, including nematodes, are common throughout the world (Nansen & Roepstorff, 1999) and cause significant economic losses to pig producers, as the nematodes may affect the overall growth rate and feed utilisation efficiency (e.g. Hale & Stewart, 1979; Hale & Marti, 1984; Hale et al., 1981, 1985; Stewart et al., 1985). In extreme cases the nematodes may also cause the death of

infected animals (e.g. Jensen & Svensmark, 1996). This problem is particularly significant for the organic pig husbandry, as a goal of organic production is to minimise or entirely eliminate the use of medical drugs, including anthelmintics, and because nematode occurrence is generally increased in organic animals systems and other alternatives to industrial husbandry systems, as these generally offer better conditions for development and survival of infective parasite stages (deep litter systems, outdoor facilities), whereby the animals are much more exposed to infection (Thamsborg & Roepstorff, 2003).

Furthermore, a long series of controlled experimental infections of pigs with *Oesophagostomum* has demonstrated that diets varying in carbohydrate source and in contents of insoluble fibre (but otherwise very similar) may influence not only the fecundity of the nematode females, but also the establishment and survival of the worms. Unfortunately, high contents of fibre and partially undegradable carbohydrates, as found in standard organic swine diets, seem to be favourable for the parasites, while parasite unfavourable diets composed of highly degradable carbohydrates are not normally fed to pigs (Bjørn et al., 1995; Petkevičius *et al.*, 1997, 1999, 2001).

There have been many alternative approaches towards new methods for parasite control, and one of the more promising is actually the manipulation of dietary composition. Recent data thus demonstrate that it is possible to identify organically relevant and economically competitive carbohydrate sources with high contents of fructooligosaccharides, on which the pigs grow well, and that these diets may reduce worm numbers and female fecundity of both *Oesophagostomum* and *Trichuris* markedly (Mejer *et al.*, unpublished; Thomsen *et al.*, unpublished). Petkevičius *et al.* (unpublished) found a markedly reduced egg excretion and an almost complete elimination of *O. dentatum* from pigs fed a diet with added inulin. Thus, novel feeding strategies that include continuous or periodical supplements of diets rich in fructooligosaccharides may contribute to future sustainable nematode control in pigs.

A pure inulin product has up till now been an expensive product and therefore probably not likely to be used as a feed supplement in commercial pig production. Though the price may decrease with increased demand in the production units there

is also the basis for an alternative product that can be produced at a competitive cost.

**Summary of invention**

The present invention relates to a method for reducing or removing off-odour and off-flavour in animals, said method comprising feeding to an entire male, castrate male and female animals a chicory root product during at least two days prior to slaughtering the animal. Preferably the animal is a domesticated animal, more preferable the animal is a pig.

Feeding animals with chicory root products reduces or removes boar taint in animals and improves the meat quality according to use of the meat as human food. The reduction of boar taint is also connected with reducing malodour in the environment of the live animals due to offensive-smelling compounds in the mixture of faeces and urine of the animals (liquid manure). A chicory root product is a cheap product and the effect of the product is more effective and efficient in reducing such taints than feeding animals with compounds such as inulin isolated from chicory plants, thus an alternative product to pure inulin is chicory roots. Also the chicory root product has beneficial effects on the animals, effects which can not be obtained by pure inulin, one of these effects are effect on meat taste.

In another embodiment the invention relates to a method for reducing the skatole content in animals, said method comprising feeding to an animal a chicory root product for at least two days prior to slaughtering.

In a further embodiment the invention relates to a method for reducing the androstenone content in meat and fat and blood said method comprising feeding to an animal a chicory root product for at least two days.

Skatole and androstenone are two of the compounds resulting in boar taint of entire male pig meat, and are connected to off-odour and flavours of meat. Reducing skatole and androstenone content in meat also decreases the amount of animals being rejected at slaughter for use in meat cuts.

In yet another embodiment the invention relates to a method for improving the odour, flavour, taste and aftertaste of meat from a human sensory acceptability perspective, said method comprising feeding to an animal a chicory root product for at least two days prior to slaughter. The chicory root product has an effect on taste and aftertaste of meat, which can not be obtained by feeding animals with pure inulin.

In a further embodiment the invention relates to a method for reducing malodour as related to the live animals environment, said method comprising feeding a chicory root product to animals for at least two days. Reducing malodour compounds coming from pig stables and manure lead to environmental benefits in relation to the public.

In another embodiment the invention relates to a method for reducing the amount of infections with pathogens of the gastrointestinal tract in a non-human animal, said method comprising feeding to a non-human animal a chicory root product for at least two days.

The invention relates to animal welfare by a friendly, humane, low cost and highly effective feeding methodology when compared to all the presently utilised methods for boar taint control.

**Brief description of Figures**

Fig. 1. (a) The scores of odorous compounds of raw data from colon contents of control-fed and chicory-fed pigs. (b) The loadings of the odorous compounds of control-fed and chicory-fed pigs.

Fig. 2. (a) The scores of low threshold values of odorous compounds from colon contents of control-fed and chicory-fed pigs. (b) The loadings of low threshold values of the odorous compounds of control-fed and chicory-fed pigs.

Fig. 3. (a) The scores of high threshold values of odorous compounds from colon contents of control-fed and chicory-fed pigs. (b) The loadings of high threshold values of the odorous compounds of control-fed and chicory-fed pigs.

Fig. 4. Mean *Oesophagostomum dentatum* egg counts (eggs per gram faeces) in five groups of eight pigs fed different diets. The first 28 days after infection with 3000 *O. dentatum* L3-larvae all pigs were given concentrate and grass silage. Thereafter the concentrate control group was given only concentrate and the long-term chicory group had the silage substituted for shredded chicory roots. This was also done for the short-term chicory group 28 days before slaughter.

Fig. 5. Principal Component Analysis (PCA) of sensory profiling data from freshly cooked entire male pork meat samples for each of four feeding treatments, 1). Non-Bioactive Control, 2). Silage, 3). Chicory, and 4). Chicory/Silage.

**Definitions:**

A chicory root product: By a chicory root product is intended first and foremost the complete chicory roots. Also fractions of chicory root are included. Also encompassed by the present invention are processed products thereof, e.g. pulp, flakes, powder, flour, dried pulp, dried flakes, dried tubers, silage, enzymatically processed products, microbiologically processed products.

A chicory root extract: An extract made from chicory roots, wherein the extract comprises an inulin and/or FOS fraction as well as a low molecular weight fraction Low molecular weight compounds are compounds below 2000 Dalton. Preferably the extract comprises the coumarins i.e. esculetin, sesquiterpenes, terpene, phytosterol, polyamine and flavonoid.

A pig: An animal belonging to the group of animals characterised by the Latin name *Sus scrofa*.

Bitter chicory: By bitter chicory is to be understood chicory with a bitter taste. Bitter chicory need not be different from chicory or chicory root product.

Boar taint: Off-odour and/or off-flavour of animal products including meat.

Domesticated animals: cattle, sheep, goat, pig, horse, donkey, dog, cat, poultry, chicken, duck, goose, turkey, steer, mink.

Pigs can be classified according to age and partly according to weight. For the purposes of the present invention the following classification is used:

| | |
|---|---|
| Suckling piglet: | 0-4 weeks or until 7 weeks of age (until weaning) |
| Weaned pigs: | 4-8 weeks of age |
| Growing pigs: | above 8 weeks. |

Growing pigs are often referred to as Porkers (50-60 kg), finishers or fatteners (both up to 160 kg).

Chicory: By a Chicory plant is intended any species, subspecies or variety, which is a member of the Genus Cichorium L. belonging to the Compositae. Some botanists place the Cichorium family in the Asteraceae. Known species include at least:

Cichorium alatum Hochst. & Steud.ex DC.
Cichorium ambiguum Schult.
Cichorium aposeris E.H.L.Krause
Cichorium arnoseris E.H.L.Krause
Cichorium balearicum Porta
Cichorium barbatum E.H.L.Krause
Cichorium bottae Deflers
Cichorium bottae Deflers
Cichorium byzantinum Clem.
Cichorium caeruleum Gilib.
Cichorium callosum Pomel
Cichorium calvum Sch.Bip.
Cichorium casnia C.B.Clarke
Cichorium cicorea Dum.
Cichorium commune Pall.
Cichorium cosnia Buch.-Ham.
Cichorium crispum Mill.
Cichorium dichotomum Link
Cichorium divaricatum Heldr.ex Nym.
Cichorium divaricatum Schousb
Cichorium dubium E.H.L.Krause

Cichorium endivia Linn.
Cichorium endivia subsp. divaricatum (Schousboe) P.D.Sell
Cichorium endivia subsp. pumilum (Jacq.) C.Jeffrey
Cichorium esculentum Salisb.
Cichorium glabratum Presl
Cichorium glandulosum Boiss. & Huet
Cichorium glaucum Hoffmgg. & Link
Cichorium hirsutum Gren.
Cichorium intybus convar. foliosum (Hegi) J.Holub
Cichorium intybus convar. radicosum (Alef.) J.Holub
Cichorium intybus forma alba Farw.
Cichorium intybus forma rubicunda Farw.
Cichorium intybus L.
Cichorium intybus Linn.
Cichorium intybus subsp. glabratum (C.Presl) G.Wagenitz & U.Bedarff
Cichorium minimum Portenschl.
Cichorium nanum Portenschl.ex Nym.
Cichorium noeanum Boiss.
Cichorium officinale Gueldenst.ex Ledeb.
Cichorium perenne Stokes
Cichorium polystachyum Pomel
Cichorium pumilum Jacq.
Cichorium rhagadiolus E.H.L.Krause
Cichorium rigidum Salisb.
Cichorium spinosum Linn.
Cichorium sylvestre Garsault
Cichorium sylvestre Lam.

A commonly used agricultural variety of Cichorium is:
Cichorium intybus L. var. Orchies

Plant varieties of Cicorium for which Plant variety protection has been granted or is about to be granted at the Community Plant Variety Office, Angers, France

**Cichorium endivia L.**

| File Number | Application Date | Denomination | Grant Number | Grant Date | End of Protection |
|---|---|---|---|---|---|
| 19971402 | 28/11/1997 | ARIGA | 3152 | 02/06/1998 | 31/12/2023 |
| 19951973 | 25/08/1995 | ATRIA | 1635 | 15/01/1997 | 31/12/2022 |
| 19950129 | 31/05/1995 | BOLDIE | 1088 | 15/10/1996 | 01/10/2017 |
| 19970359 | 12/03/1997 | BOOGIE | 3047 | 06/07/1998 | 31/12/2023 |
| 19980139 | 23/01/1998 | CENTURY | 3605 | 19/10/1998 | 31/12/2023 |
| 19990460 | 22/03/1999 | EMILIE | 7833 | 11/06/2001 | 31/12/2026 |
| 19950621 | 09/08/1995 | EXCEL | 1459 | 16/12/1996 | 31/12/2021 |
| 19970357 | 12/03/1997 | FOXIE | 3134 | 02/06/1998 | 31/12/2023 |
| 19971458 | 15/12/1997 | FREHEL | 5639 | 20/12/1999 | 31/12/2024 |
| 20001725 | 21/11/2000 | GIRONA | 9370 | 06/05/2002 | 31/12/2027 |
| 20001830 | 06/12/2000 | ISADORA | 7998 | 06/08/2001 | 31/12/2026 |
| 20001831 | 06/12/2000 | ISOLA | 7999 | 06/08/2001 | 31/12/2026 |
| 19990309 | 01/03/1999 | KETHEL | 7446 | 09/04/2001 | 31/12/2026 |
| 19991249 | 08/09/1999 | KIBRIS | 10192 | 21/10/2002 | 31/12/2027 |
| 20000439 | 23/03/2000 | LASSIE | 8505 | 03/12/2001 | 31/12/2026 |
| 20000809 | 31/05/2000 | LILIE | 7390 | 05/03/2001 | 31/12/2026 |
| 19950622 | 09/08/1995 | MISTRAL | 1460 | 16/12/1996 | 31/12/2021 |
| 19991604 | 15/11/1999 | MONTREAL | 8500 | 03/12/2001 | 31/12/2026 |
| 19950623 | 09/08/1995 | NAOMI | 1461 | 16/12/1996 | 31/12/2021 |
| 19951225 | 29/08/1995 | NATACHA | 1089 | 15/10/1996 | 01/02/2018 |
| 19950294 | 27/04/1995 | NUANCE | 975 | 02/09/1996 | 01/09/2016 |
| 20001829 | 06/12/2000 | OLIVIA | 7997 | 06/08/2001 | 31/12/2026 |
| 19951972 | 25/08/1995 | PRADA | 1634 | 15/01/1997 | 01/10/2027 |
| 19971403 | 28/11/1997 | SACHA | 3151 | 02/06/1998 | 31/12/2023 |
| 19950258 | 06/07/1995 | SARDANA | 1942 | 15/05/1997 | 01/06/2017 |
| 19981452 | 29/10/1998 | SNOOPIE | 5566 | 06/12/1999 | 31/12/2024 |
| 19991208 | 27/08/1999 | STOMIE | 5801 | 14/02/2000 | 31/12/2025 |
| 19970360 | 12/03/1997 | TRUDIE | 3048 | 06/07/1998 | 31/12/2023 |
| 19970358 | 12/03/1997 | WOODIE | 3132 | 02/06/1998 | 31/12/2023 |

**Cichorium endivia L.**

| File Number | Application Date | Breeder's Reference | Proposed Denomination |
|---|---|---|---|
| 2001/0741 | 25/04/2001 | e 02 2216 | ATLETA |
| 2000/1908 | 10/01/2001 | bejo 1978 | CARLOS |
| 2002/1355 | 30/10/2002 | 11-122 rz | CASAL |
| 2000/1911 | 10/01/2001 | bejo 1895 | DAVOS |
| 2002/1356 | 30/10/2002 | 11-510 rz | LASKO |
| 2000/1907 | 10/01/2001 | bejo 1979 | LEXOS |
| 2002/1354 | 03/09/2002 | 11-194 rz | MARCONI |
| 2000/1910 | 10/01/2001 | bejo 1894 | MONOS |

**Cichorium intybus L. partim**

| File Number | Application Date | Denomination | Grant Number | Grant Date | End of Protection |
|---|---|---|---|---|---|
| 19980340 | 11/03/1998 | AUG133 | 7466 | 09/04/2001 | 31/12/2026 |
| 19980339 | 11/03/1998 | CES4731 | 7465 | 09/04/2001 | 31/12/2026 |
| 19970157 | 28/01/1997 | CPZ 4641 | 6897 | 20/11/2000 | 31/12/2025 |
| 19970166 | 28/01/1997 | CPZ 6722 | 6898 | 20/11/2000 | 31/12/2025 |
| 19952585 | 24/08/1995 | CRP 308-2 | 1464 | 16/12/1996 | 31/12/2021 |
| 19952584 | 24/08/1995 | CRP 609-3 | 1463 | 16/12/1996 | 31/12/2021 |
| 19980990 | 17/07/1998 | FASTE | 7472 | 09/04/2001 | 31/12/2026 |
| 19960978 | 03/09/1996 | FLA A1-1 | 2444 | 01/09/1997 | 31/12/2022 |
| 19960977 | 03/09/1996 | FRAN B1-2 | 2443 | 01/09/1997 | 31/12/2022 |
| 19980991 | 17/07/1998 | OESIA | 5844 | 03/04/2000 | 31/12/2025 |
| 19970156 | 28/01/1997 | SISTA153 | 6896 | 20/11/2000 | 31/12/2025 |

**Cichorium intybus L. partim**

| File | Application | Breeder's | Proposed |
|---|---|---|---|

| Number | Date | Reference | Denomination |
|---|---|---|---|
| 2001/1199 | 19/07/2001 | bejo 2202 | FOX 14 |
| 2000/1913 | 10/01/2001 | bejo 2196 | FURB 21 |
| 2000/1909 | 10/01/2001 | bejo 2197 | NER261 |
| 2002/0434 | 19/03/2002 | nun 9001 cm | NUN9001CM |
| 2002/0435 | 19/03/2002 | nun 9002 cm | NUN9002CM |
| 2002/0436 | 19/03/2002 | nun 9003 cm | NUN9003CM |
| 2002/0437 | 19/03/2002 | nun 9004 cm | NUN9004CM |
| 2002/0438 | 19/03/2002 | nun 9005 cm | NUN9005CM |
| 2002/0439 | 19/03/2002 | nun 9006 cm | NUN9006CM |
| 2002/0440 | 19/03/2002 | nun 9007 cm | NUN9007CM |
| 2000/0303 | 28/02/2000 | e 84.025 | REDORIA |
| 2000/1914 | 10/01/2001 | bejo 2195 | SISTA 159 |
| 2001/0740 | 25/04/2001 | wo118 | WO 118 |
| 1999/0819 | 08/06/1999 | wo 125 | WO125 |
| 1999/0818 | 08/06/1999 | wo 126 | WO126 |

**Detailed description of the invention**

The present invention relates to a method for reducing taint in animals, said method comprising feeding to an animal a chicory root product during at least one day preferably at least two days prior to slaughtering the animal. The taint is connected to malodour in places where animals are living especially in indoor locations e.g. in stables, other houses or hiding-places for pigs. The taint is also connected to off-odour and flavour in meat from a human sensory perspective.

By using the wording 'reducing taint in animals' it is not meant to limit the reduction of taint to the inside of the animals e.g. in all food related items contained in the animal in particular the meat, also the stables and outdoor areas where animals are living are intended to be included as well as the manure/slurry kept in tanks and spread on the soil. In general the reduction of taint in the environment of animals is included.

Feeding the animals with the chicory root product reduces taint in animals, males as well as females. Surprisingly the effect of the chicory root product on skatole in backfat is higher than expected when comparing to results of an experiment using purified inulin (Claus, 1992 & 1994).

Feeding male and female animals with chicory root product reduces the off odour and off flavour of the meat and hereby increases the human sensory enjoyment in eating the untainted meat. The reduction of off odour and off flavour also reduces the amount of animals that are being degraded as boar tainted meat discharged due to unsuitability to be used directly as a human food.

Furthermore, castration of the animals can be avoided, which increase the animal welfare due to avoiding the pain male animals are subjected to at the time of castration. The chicory root product is a cheap alternative to castration especially in countries where authorised veterinarians perform the castration.

**Feeding time**

The chicory root product can be produced from plants of one or more of the species, genus or plant families mentioned above. This chicory root product is fed to the animal for at least 1 day, such as at least 2 days, such as at least 3 days, such as at least 4 days, such as at least 5 days, such as at least 6 days, such as at least one week, for example at least 1.5 weeks, such as at least 2 weeks, preferably at least 3 weeks, such as at least 4 weeks, for example at least 5 weeks, such as at least 6 weeks, for example at least 7 weeks, such as at least 8 weeks, for example at least 9 weeks, such as at least 10 weeks, for example at least 15 weeks, such as at least 20 weeks.

Feeding animals with the chicory root product within a short period just prior to slaughtering reduces the amount of chicory root product to be used, simultaneously with reducing taint of the meat of the animal. Feeding animals with the chicory root product within a long period prior to slaughtering generally reduces taint of the meat and malodour of the stables or living place of the animal as well as the parasite load.

To reduce taint in the animal the chicory root product is fed to the animal substantially until slaughter. The initiation of the feeding by the chicory root product can be at any time during the life of the animal. The amount of chicory root product per kg animal eaten by said animal may vary during the life of the animal or during the year due to the need of the chicory root product or due to fluctuation in the quality of the feed, where the quality can be of the chicory plants or the other feed components.

In the period where the animal is fed by the chicory root product as outlined elsewhere, the chicory root product is fed to the animal daily. The frequency of daily feeding may vary from one portion which is eaten up by the animal within a short period or the animal can have admission to the chicory root product all day long, preferred is that the chicory root product is fed to the animal several times daily, such as 2 times, 3 times, 4 times, 5 times, or more than 5 times.
The chicory root product can be fed to the animals every day, every second day, every third day, every fourth day, every fifth day, every sixth day or one a week. Preferred is when the animals are fed by the chicory product every day.

The animals may also be allowed to crop an area with growing chicory plants, hereby the animals can eat the leaves of the plants and/or eat the roots by first digging up or drawing up the plants and/or roots.

The animals may also crop an area where chicory plants are harvested. The animals can eat the remaining chicory plants or the remaining chicory plant parts. Remaining plant parts can be due to topping the plants when harvesting, removing the roots and leaving the leaf part on the area. Also non-removed roots can be eaten by the animal.

**Feed ration**

The chicory root product can constitute a part of the daily feed ration, preferably the chicory root product part of the ration of the animal is at least 5 % on a daily energy basis. Also preferred is when the chicory root product part of the ration of the animal is at least 10% on a daily energy basis.

Further, when feeding with the chicory root product the ration based on a daily energy basis can be that the chicory root product part comprises at least 15 % of the ration, more preferably at least 20%, more preferably at least 25%, more preferably at least 30 %, for example at least 35%, such as at least 40%, for example at least 50%, such as at least 60%, for example at least 75%, such as at least 90%, for example substantially 100%.

The chicory root product does not seem to result in a reduction in the growth rate of the animals; furthermore the animals do not show signs of avoiding eating the chicory root product.

The chicory root product is a protein-free or substantially protein-free product. Surprisingly, when the animals are fed with the chicory root product, the animals need not be fed with an additional protein supplying product to obtain the weight of an animal fed by ordinary feeding products.

**Animals**

In one embodiment the animal as described herein may be any higher animals at any stage of life, preferable the animals is domesticated animals and more preferred is when the animal is a ruminant, such as cow, sheep, goat, buffalo, deer, cattle, antelope.

In another preferred embodiment the animal is a monogastric species, such as horse, pig, poultry, dog, cat. Further preferred is when the monogastric species is a pig. Yet further preferred is when the pig is an entire male pig.

Piglets can eat the chicory root product from none, one or several days before weaning from the sow as a part of the ration as described elsewhere. Preferred is feeding pigs with the chicory root product wherein weight of the pig is from 25 to 300 kg, more preferably as from 55 to 130 kg, which is the weight of fatteners at slaughtering. Pigs of all ages can be feed with the chicory root product such as to suckling piglet of 0-4 weeks of age (or until weaning), weaned pigs of 4-8 weeks of age, growing pigs above 8 weeks for instance is growing pigs often referred to as porkers (50-60 kg), finishers or fatteners (both up to 130 kg). The pigs can be feed

with the chicory root product when the pigs are ranging in weight from 4 to 350 kg, such as 5 to 150 kg, e.g. such as 5 to 30 kg, further such as 30 to 50 kg, such as 50 to 80 kg, such as 80 to 110 kg, such as 110 to 140 kg, such as 140 to 170 kg, such as 170 to 200 kg, such as 200 to 275 kg, such as 275 to 350 kg.

The animals fed by the chicory root product of the invention may live in organic or non-organic production systems. The animals may be in a stable all day or have access to outdoor equipment such as a fence.

**Chicory plants**

The chicory root product described herein can be prepared from plants of the family *Compositae*, the chicory root product can be produced from plants of one or more genus of the family *Compositae*, preferred is plants from the genus *Cichorium*. In this context chicory is used to describe plants belonging to the genus *Cichorium*. As just mentioned the plants may belong to a single or more genus of family *Compositae* as well as from a single or more species of the genus *Cichorium*, as well as from a single or more varieties of the species *Cichorium intybus L.* Preferably the plants are of the species *Cichorium intybus L.* The genera and species referred to are that mentioned previously. The varieties are any chicory variety, which are being cultivated at a time. Preferred are plants of agricultural varieties. More preferred are plants with large roots, most preferred are varieties with a high biomass yield by area e.g. 60 ton per ha. Further preferred are varieties with a large inulin content, such as at least 15% inulin on a dry matter basis, e.g. at least 20% inulin, such as at least 30% inulin, e.g. at least 40% inulin, such as at least 50% inulin, for instance at least 60% inulin, such as at least 70% inulin, for instance at least 80% inulin, such as at least 90% inulin, for instance at least 95% inulin.

Chicory plants are easy to grow and many agriculture varieties have a high yield, hereby the chicory root product becomes a cheap product. Furthermore the chicory plants can be handled by equipment used in sugar beet production.

All parts of the chicory plant can be used to prepare a chicory root product; the phrase 'chicory root product' is used to indicate that preferably the main part of the

product is prepared from the roots of the chicory plants. This root part of the amount of chicory plant used to produce the chicory root product may constitute more than 20% of the total dry weight of chicory plant used, such as more than 30%, such as more than 40%, such as more than 50%, such as more than 60%, such as more than 70%, such as more than 80%, such as more than 90%, such as substantially 100%.

In the description of the chicory roots e.g. characterisation of the contents of compounds these characteristics may be valid for portions including entire plants or portions only including roots and small parts of the leafs.

The chicory root product is prepared from chicory plants, wherein the chicory roots contain at least 5% inulin, more preferably at least 10% inulin, more preferably at least 15 % inulin, more preferably at least 20 % inulin, such as at least 25% inulin, for example at least 30 % inulin on wet weight basis of the root.

The chicory root product is prepared from chicory plants, wherein the chicory roots contain at least 5% FOS, more preferably at least 10% FOS, more preferably at least 15 % FOS, more preferably at least 20 % FOS, such as at least 25% FOS, for example at least 30 % FOS on wet weight basis of the root. FOS is fructooligosaccharides.

**Processed chicory root products**

The chicory root product used according to this invention can be a processed chicory root product comprising a silage product of chicory roots, such as a silage product of essentially whole chicory roots.

**Silage**

Silage is prepared by anaerobic fermentation this can be in a pit, silo or other enclosure or by chemical preservation e.g. by lactic acid, propionic acid, and formaldehyde. The chicory plant parts or chicory roots can be ensiled alone meaning without other plant species or it can be ensiled together with different plant species of forage crops such as ryegrass, maize, sorghum, alfalfa, potatoes, beets e.g. sugar beets.

The plant material is harvested green and stored as fresh material, enclosed in air-proof conditions (pit, or under a plastic or similar covering) and allowed to ferment, with most of the soluble sugars converted to low molecular weight volatile fatty acids, such as acetic acid. Various additives may be used, either to increase the concentration of fermentable carbohydrate (molasses), to increase the proportion of beneficial bacteria e.g. lactic acid in the ensiled material, or to artificially lower the pH of the mixture. Additional fermentable carbohydrate may be added as molasses. Alternatively, enzymes such as xylanases and cellulases may be added to release low molecular weight fermentable substrates from the cell wall polysaccharides. Synthetic volatile fatty acids e.g. propionic acid may also be added to lower pH.

The chicory root silage can also be produced by mixtures of chicory plants and straw or by adding pellets from sugar beet pulp to the chicory plants. Other dried products can also be used e.g. potato starch.

The silage can constitute the chicory root product or the chicory root product can be produced from silage and other chicory products described herein.

**Fermented product**

The chicory root product of the invention can be a product, wherein the chicory root product comprises a fermented product of chicory roots. The fermentation can be initiated with fractions of fresh roots, fractions of dried roots and extracts.

A fermented chicory root product can be obtained by fermentation with bacteria such as *Bacillus*, *Acetobacter*, etc also yeast can be used to the fermentation process. Preferred is fermentation with *Lactobacillus casei alactosus*, *Lactobacillus cellobiosus*, *Leuconostoc destranicum*, *Leuconostoc mesenteroids* , *Streptococcus lactis*, *Streptococcus diacelylactis*, *Saccharomyces florentinus*. Methods of fermentation of chicory roots are described in US 4,671,962.

**Decomposition of chicory roots**

Heating and/or drying chopped chicory roots may carry out decomposition of the chicory roots. Furthermore, the decomposition may be performed by first chopping

and grinding the chicory roots into fine pieces, then preparing a slurry of the pieces, and enzymatically decomposing the slurry; or alternatively by first chopping the chicory roots into fine pieces, then heating and drying them, adding thereto water to form a slurry, and enzymatically decomposing the slurry.

If necessary, a further treatment may be conducted by the use of pectinase and/or cellulase. Afterward, an endo-type inulase is added to the slurry, and the enzymatic decomposition is then performed at a temperature of 40°C to 80°C for 12 to 36 hours. Preferred is a product in which 50 weight % or more of the solids content comprises the fructooligosaccharide (FOS).

Usable examples of the endo-type insulase include enzymes produced by mold fungi such as those of the genus *Aspergillus* (*A. niger* and the like) and those of the genus *Penicillium* (*P. trzebinskii* and the like), and bacteria such as *Bacillus* (*B. circulans* and the like). In a preferable case, the endo-type inulase wherein the optimum temperature is from 30°C to 80°C and the optimum pH is from 4 to 7 is used, so that the oligosaccharide is effectively produced from the chicory flakes. In the practical enzyme decomposition, it is preferable that the temperature of the enzymatic decomposition is high for the sake of preventing contamination with various bacteria. Therefore, the enzymatic decomposition is suitably performed at a temperature of 40° to 80°C. Enzymatic preparation is further described in US 4,971,815.

The chicory root product of the invention can be a product, wherein the chicory root product comprises flour of chicory root. This invention therefore provides a process for the preparation of a flour from tubers of chicory or similar inulin-containing plants, which process comprises the steps of: (a) macerating the tubers to a homogenate; (b) heating the homogenate at a temperature ranging from about 150°C to about 90°C for a time ranging, respectively from about 15 seconds to about 10 minutes; (c) subjecting the heated homogenate to spray drying in a stream of hot gas; and (d) recovering a flour comprising a mixture of monosaccharides, small oligosaccharides and large oligosaccharides. Flour production is further described in US 4,871,574.

The chicory root product of the invention can be a product, wherein the chicory root product comprises pulp of chicory root. Suitable pulps include those where some of the inulin has been removed (extracted) to leave a chicory pulp. The present inven-

tion includes all chicory pulp, which can be obtained from chicory plants, including the whole range of possible fibre and inulin content. The pulp is preferably obtained from at least chicory root material. The chicory pulp may be incorporated into a chicory root product with the same composition as directly produced from the extraction procedure. Alternatively, the pulp may undergo one or more steps to obtain a pulp of a different composition and/or form. For example, the pulp may be dried and then ground up to provide a dry product of small particle size, which may be used to produce a chicory root product.

The chicory root product of the invention can be a product, wherein the chicory root product comprises a dried product of chicory roots, such as a dried product of essentially whole chicory roots. The chicory roots or disintegrated chicory roots can be dried by any drying method, such as sun dried, dried by heat, dried by air, dried by heated air, dried in a heating chamber or freeze dried

The chicory root product of the invention can be a product, wherein the chicory root product is a disintegrated product, such as a powder, flakes, pulp, slices, flour, and pellets. The chicory root product can be disintegrated before a possible production process or the processed chicory root product can be disintegrated at a stage within the processing steps or followed processing. One example of drying of chopped chicory roots is at 60°C for 3 days in a heating chamber, which results in 3-4 % water content. The chicory roots can be homogenised, cut into strips, planed or disintegrated in other ways.

The chicory root product of the invention can be a product, wherein the chicory root product comprises fresh chicory roots. By fresh is meant a period of time from the chicory plants has been harvested to some months of storage such as 1 month, e.g. 2 months, such as 3 months, such as 4 months, such as 5 months such as 6 months, such as 7 months, such as 8 months, such as 9 months, such as 10 months, such as 11 months, such as 12 months. At the storage period the chicory roots can be stored at options where the roots do not ensilage, and/or ferment and/or dry. Some of the roots within the storage pile can locally ensilage, ferment or dry, which is accepted. One storage option is to collect the chicory roots in heaps or piles at conditions preventing silage formation, fermentation or drying. A certain

degree of drying is acceptable, such as loss of 50% of the water content of the freshly harvested chicory roots.

## Fractions of chicory roots

The chicory root product of the invention can be a product, wherein the chicory root product comprises a fraction and/or an extract of chicory roots. The fraction of the chicory root product comprises inulin and oligofructose and at least one other compound from the chicory roots.

As mentioned elsewhere the chicory root product need not only to be produced from chicory roots or parts of chicory plants. To produce a chicory root product fraction and/or extract of chicory roots can be added to other feeding components.

Extract can be produced by extraction of compounds in an aqueous mixture of disintegrated chicory roots and a liquid or in a mixture of different liquids. The disintegrated chicory roots are described above.

The fraction and/or extract of chicory root preferably comprise inulin and oligofructose fractions and a low molecular weight fraction comprising coumarins and/or sesquiterpenes. The fraction and/or extract of chicory root can also comprise other secondary metabolites as mentioned below.

## Secondary metabolites

Secondary metabolites are compounds which are not a part of the primary metabolism of the organism e.g. they are not amino acids, carbohydrates, lipids and nucleic acids. The secondary metabolites in chicory can be divided in several chemical classes: terpenes, phytosterols, polyamines, coumarins and flavonoids. The content of secondary metabolites in a plant can vary according to season, growth conditions, variety, anatomical part of the plant, age of the plant and degree of attack of insects, herbivores or plant diseases e.g. bacteria or fungi.

Preferred secondary metabolites of chicory root fractions of the invention are selected from the groups mentioned in the following paragraphs:

Terpenes: *Sesquiterpene lactones:* 8-Deoxylactucin, crepidiaside, lactucin, lactupicrin, crepidraside, 11-□-13-dihydrolactucin, picriside, sonchuside A, sonchuside C, cichoriolide A, cichoriosides A, cichorioside B and cichorioside C.

*Phytosterols:* Sitosterol, stigmasterol, and campersterol.

Coumarines: Esculetin (=aesculetin), esculin (the glucon of esculetin), cichoriin-6'-p-hydroxyphenyl acetate and cichoriin.

Flavonoids: Luteolin 7-glucuronide, quercetin 3-galactoside, quercetin 3-glucuronide, kaempferol 3-glucoside, kaempferol 3-glucuronide, isorhamnetin 3-glucuronide.

*Anthocyanins:* Cyanidin 3-O-□-(6-o-malonyl)-D-glucopyranoside and four delphinidin derivatives.

Caffeic acid derivatives: Caffeic acid, chicoric acid, and chlorogenic acid.

Polyamines (biogenic amines): Putrescine, spermidine, spermine.

More preferred is secondary metabolites selected from the groups of terpenes, coumarines and caffeic acid derivatives. The most preferred secondary metabolites from these groups comprises:
Terpenes: *Sesquiterpene lactones:* 8-Deoxylactucin, crepidiaside?, lactucin, lactupicrin, crepidraside, 11-□-13-dihydrolactucin, picriside, sonchuside A, sonchuside C, cichoriolide A, cichoriosides A, cichorioside B and cichorioside C.
Coumarines: Cichoriin-6'-p-hydroxyphenyl acetate, Esculetin (=aesculetin), and esculin.
Caffeic acid derivatives: Caffeic acid, and chicoric acid.

**Skatole**

Another aspect of the invention is a method for reducing the skatole content in animals, said method comprising feeding to a animal a chicory root product for at least two days prior to slaughtering. With regard to this aspect, it can be combined

with the characteristics described above, especially in condition to feeding of animal and production of chicory root product.

By feeding animals with the chicory feed product the skatole content of blood plasma is reduced by at least 25%, more preferably at least 40%, more preferably at least 50%, more preferably at least 75%, more preferably at least 80%, more preferably at least 90%, more preferably at least 95%, more preferably at least 98%, more preferably to substantially 0. Surprisingly the reduction of skatole in the blood plasma is greater than expected.

It is preferred that the method of feeding animals with chicory root product is one wherein the skatole content of blood and/or fat is reduced to below the unacceptable human off odour and flavour sensory threshold and maybe even to zero in meat produced from the animals, this is of additional importance as skatole functions as an enhancer of the sensory off-odour/flavour producer androstenone and maybe other off odour/flavour components of unknown origin.

Preferred is that the skatole content of backfat and/or meat is reduced by at least 25%, more preferably at least 40%, more preferably at least 50%, more preferably at least 75%, more preferably at least 80%, more preferably at least 90%, more preferably at least 95%, more preferably at least 98%, more preferably to substantially 0.

Also preferred is that the skatole content of manure is reduced. Skatole in manure can be picked up through the skin when the animals lie in or roll in the manure Hansen et al., 1994). Some animals lie in manure to be cooled in the summer, this especially concerns pigs. By this contact between skin and manure the skatole is absorbed through the skin and further transported to the blood, fat and meat. In this way the skatole content in animals can be too high and influence the meat quality e.g. boar taint. Also female and castrated male pigs can obtain a too high content of skatole in the blood, fat and meat e.g. by uptake through the skin (Hansen et al., 1994 & 1995). Reduction of skatole content of backfat and meat of female and castrated male pigs are preferred.

The chicory root product also has an effect on female and castrated male animals resulting in a reduction of skatole content of blood, fat and meat too.

**Androstenone**

The inventors have surprisingly discovered that the amount of androstenone in the blood might be significant lowered by feeding animals with the chicory root product, thus another aspect of the invention is a method for reducing the androstenone content in meat and/or fat and/or blood said method comprising feeding to an animal a chicory root product for at least two days.

Preferred is that the androstenone content is reduced by at least 10%, more preferably at least 25%, more preferably at least 40%, more preferably at least 50%, more preferably at least 75%, more preferably at least 80%, more preferably at least 90%, more preferably at least 95%, more preferably at least 98%.

Preferred is that the androstenone content in blood, meat and/or fat is reduced to below the human off odour/flavour sensory threshold around 1.0 ppm in backfat. However, the off odour/flavour sensory threshold is different from one person to another. Furthermore the off odour/flavour sensory threshold is unknown when skatole concentration is nearly zero.

It is further preferred that the method as described is used until the animal is subsequently slaughtered.

The aspect of the invention comprising a method for reducing the androstenone content in meat, and/or fat and/or blood can be combined with any characteristic of animal and chicory root product as described elsewhere herein.

**Sensory characteristics**

Another aspect of the invention is a method for improving the odour, flavour, taste and aftertaste of meat from a human sensory perspective in relation, said method comprising feeding to an animal a chicory root product for at least two days prior to slaughter.

The improvement of sensory characteristics comprises reduction of sensory characteristics classified as: Piggy/Animaly-odour, Meat/Gamey-odour, Cardboard-odour, Linseed oil-odour, Sweet-taste, Salt-taste, Piggy/Animaly-flavour, Cooked ham-flavour, Fresh cooked pork meat-flavour, Cardboard-flavour, White pepper-flavour, Pork fat-flavour, Lactic/Fresh sour-aftertaste, spicy heat aftertaste, Chemical/medicinal-aftertaste, Fatty mouth coating-aftertaste, Unacceptable.

Also the improvement of sensory characteristics comprises increasing the relative levels of the sensory characteristics classified as: Fresh cooked pork meat like-odour, Sour-taste, Bitter-taste, Metallic-flavour, Meat/Gamey-flavour, Herby-flavour, Spicy-flavour, Lactic/fresh sour-flavour, Astringent-aftertaste, Acceptable.

Reduction of malodour is of interest in production of animal wherein the animal is a ruminant such as cattle, buffalo, sheep, and goat.

Also improving the odour, flavour, taste and aftertaste of meat is of interest in animal production where the animal is a monogastric species.

It is further preferred that the monogastric animal is an animal used for meat, such as pig, poultry, rabbit, hare, more preferably wherein the monogastric animal is a pig.

The aspect of the invention comprising a method for improving the sensory characteristics as defined above in odour, taste and flavour and aftertaste of meat from a human sensory perspective can be combined with any characteristic of animal and chicory root product as described elsewhere herein.

**Stable malodour**

Another aspect of the invention is a method for reducing malodour, said method comprising feeding a chicory root product to animals for at least two days.

Reduction of malodour can be caused by a relative reduction in skatole and/or p-cresole and/or indole in the gastrointestinal tract of the animal.

Reduction of malodour directed to the environment especially in areas where humans are living has been performed by different methods as mentioned above. With the chicory root product as food for the animals, the reduction of malodour is obtained by elimination of the problem at the source, that is by avoiding the production of the offensive-smelling compounds or reducing the amount of said compounds to a level, which is not perceived as a malodour by humans. Hereby expensive equipment to reduce the malodour from the air e.g. from stables before emission to the surroundings, can be avoided.

Reduction of malodour is caused by a relative increase in the amount of 2-pentanon and/or ethylbutyrate and/or propylpropionate and/or propylbutyrate and/or butanoic acid 2-methyl-ethylester in the gastrointestinal tract of the animal.

Reduction of malodour is of interest in production of animal wherein the animal is a ruminant such as cattle, buffalo, sheep, and goat.

Also reduction of malodour is of interest in animal production where the animal is a monogastric species.

It is preferred that the monogastric animal is a furred animal, such as mink, fox, muskrat, rabbit, hare, wolf.

It is further preferred that the monogastric animal is an animal used for meat, such as pig, poultry, rabbit, hare, more preferably wherein the monogastric animal is a pig.

Reduction of malodour can occur within the animal with different influences on the surroundings. The surrounding is most influenced when the animal is indoors, according to the invention preferred is wherein the malodour is stable malodour and the animal is kept in a stable. Preferred is when the malodour is manure malodour and the manure originates from animals fed with the chicory root product.

Reducing malodour in manure influences both the conditions in stables and outdoors. When manure is collected and stored e.g. in slurry tank, until it can be

spread on land or field, malodour from the slurry tank is possible, also when spreading the manure or slurry on the fields malodour often occurs. Feeding the animal with the chicory root product reduces these malodour problems.

The aspect of the invention relating to a method for reducing malodour can be combined with any characteristic of animal and chicory root product as described elsewhere herein.

**Infections**

Another aspect of the invention is a method for reducing the amount of infections of the gastrointestinal tract in a non-human animal, said method comprising feeding to a non-human animal a chicory root product for at least two days.

Reducing infections of animals is an un-expected effect of the chicory root product, and it reduces the need for administering medicines to the animals such as anthelmintics. This is especially important in organic production systems. Both in organic and non-organic production systems the use of chicory root product as feed will increase animal welfare. The chicory root product is a cheap alternative to the medicines.

Preferred is a method for reducing the amount of infections of the gastrointestinal tract, where the infections are parasites.

Further preferred is a method for reducing the amount of infections of the gastrointestinal tract, where the parasites are worms.

One way of measuring reduction of infections is when the reduction is a reduction of the number of eggs in the animal faeces.

Preferred is reducing the amount of infections where the infections are microbiological infections selected from Coli, Salmonella, Campylobacter and Yersinia.

Further preferred is reducing the amount of infections where the infections are nematode infections selected from *Ascaris suum, Oesophagostomum dentatum, Oesophagostomum quadrispinulatum, Oesophagostomum brevicaudum, Oesophagostomum granatensis, Oesophagostomum georgianum, Hyostrongylus rubidus, Trichuris suis,* and *Strongyloides ransomi* and *Trichinella* sp.

The aspect of the invention comprising a method for reducing the amount of infections of the gastrointestinal tract in a non-human animal can be combined with any characteristic of animal and chicory root product as described elsewhere herein.

Another aspect of the invention is the use of chicory roots as a feed product for "grown up" (>> 7 weeks) pigs.

The aspect of the invention comprising use of chicory roots as a feed product for "grown up" pigs can be combined with any characteristic chicory root product as described elsewhere herein.

Another aspect of the invention is a use of chicory roots for preparing a feed product for "grown up" pigs.

The aspect of the invention comprising use of chicory roots as a feed product for "grown up" pigs can be combined with any characteristic chicory root product as described elsewhere herein.

Another aspect of the invention is a use of chicory roots for preparing a product for the prevention of boar taint.

The aspect of the invention comprising use of chicory roots for preparing a product for the prevention of boar taint can be combined with any characteristic chicory root product as described elsewhere herein.

Another aspect of the invention is a use of chicory roots for preparing a product for reduction of skatole content in pigs, in particular in boar fat.

The aspect of the invention comprising use of chicory roots for preparing a product for reduction of skatole content in pigs can be combined with any characteristic chicory root product as described elsewhere herein.

Another aspect of the invention is a use of chicory roots for preparing a product for reduction of androstenone in pigs.

The aspect of the invention comprising use of chicory roots for preparing a product for reduction of androstenone in pigs can be combined with any characteristic chicory root product as described elsewhere herein.

Another aspect of the invention is a use of chicory roots for preparing a product for reduction or prevention of gastrointestinal tract infections in pigs.

The aspect of the invention comprising use of chicory roots for preparing a product for reduction or prevention of gastrointestinal tract infections in pigs can be combined with any characteristic chicory root product as described elsewhere herein.

## Examples

### Example 1

### Feeding with chicory roots reduces the amount of odorous compounds in colon contents of pigs

Alcohols and carboxylic acids are compounds with relatively negative odour impressions. When alcohols and carboxylic acids react, pleasant smelling esters are created and the result can be a less offensive odour impact. This can be illustrated by the reaction between ethanol and butyric acid, which results in ethylbutyrate, or by the reaction between propanol and butyric acid, which results in propylbutyrate.

**Animals and feed**

The inulin content of chicory roots (variety Orchies) for the pig odour experiment was 15% on wet basis and the content of feed units for pigs was 27 FUp (pigs) per 100-kg chicory roots measured by chemical analysis in November 2001. The

experiment is a subset of an experiment, which consisted of 4 treatments each of eight pigs. The 32 pigs (16 intact male and 16 female pigs) were kept in litters of 8 pigs and fed 100 % organic concentrate and semi ad libitum grass silage until 10 December. From 10 December, the 32 pigs were distributed to the four treatments according to litter and sex in individual pens. Treatment 1 and 3 were selected for the present odour study as they represented the extremes of the treatments (Table 3). Treatment 1 was a (conventional) control group given 100-energy % organic concentrate and no roughage from 10 December until slaughter. Composition of the organic concentrate diet during the whole experiment was (g/kg): 145.5 rapeseed cake, 240.0 peas organic, 223.0 wheat organic, 220 barley organic, 50 oat organic, 100.0 GMO-free toasted soybeans, 2 Sv.vit-411 organic, 3.75 salt, 12 limestone and 3.63 monocalcium phosphate. The concentrate diet contained 8.57 MJ net energy (1.11 feed units (FUp)) and 149,7 g digestible protein per kg food. The 25 % blended organic chicory roots on energy basis plus 70 % organic concentrate were given from 10 December until slaughter of treatment 3.

Finally, 16 male pigs were slaughtered 11th and 16 female pigs 13th February 2002 for measuring meat and eating quality as well as parasites. The pigs ate the high amount of raw and bitter blended chicory roots without problems after one week of adaptation by giving individually increasing amounts of chicory roots during the first week.

The raw GC-MS areas in Table 1 and Figure 1 show that feeding pigs with the inulin containing chicory roots the fermentation pattern in the colon is shifted from protein fermentation to carbohydrate fermentation. The result is a change in composition of odorous compounds from the obnoxious protein fermentation products as p-cresol and skatole to the less offensive esters. The PCA-plot also confirms that the fermentation product pattern is well separated and mostly controlled by p-cresol and butyric acid.

**Table 1. GC-MS areas of selected compounds from the colon in chicory roots and control fed fattening pigs.**

| Treatment | 1 | | 3 | | | 1/3 |
|---|---|---|---|---|---|---|
| No. of pigs | 8 | | 7 | | | |
| Food components | 100 % organic concentrate | | 70 % organic concentrate plus 25 % chicory roots | | Significant difference between treatments | Factorial ratio between treatments |
| Compound: | LSMEAN | Std.err. | LSMEAN | Std.err. | P-value | |
| Dimethylsulfide | 83736 | 6827 | 48145 | 8078 | NS | 1.74 |
| 2-Butanon | 54274 | 7681 | 59512 | 9088 | NS | 0.91 |
| Acetic acid | 252338 | 42504 | 286741 | 50292 | NS | 0.88 |
| **2-Pentanon** | **22742** | **7513** | **48500** | **8889** | * | **0.47** |
| Dimethyldisulfide | 132354 | 52309 | 128911 | 61893 | NS | 1.03 |
| 1-Pentanol | 29277 | 6201 | 47543 | 7337 | NS | 0.62 |
| 2-Methylpropanoic acid | 43571 | 9416 | 29886 | 11141 | NS | 1.46 |
| *Ethylbutyrate (ester)* | *5026* | *28026* | *48440* | *33161* | *NS* | *0.10* |
| ***Propylpropionate (ester)*** | ***23718*** | ***40419*** | ***174429*** | ***47824*** | *(*)* | ***0.14*** |
| Butanoic acid | 935596 | 118921 | 878861 | 140710 | NS | 1.06 |
| ***Butanoic acid, 2-methyl-,ethylester*** | ***2663*** | ***1599*** | ***8679*** | ***1892*** | * | ***0.31*** |
| ***Propylbutyrat (ester)*** | ***3208*** | ***1145*** | ***7760*** | ***1355*** | * | ***0.41*** |
| 3-Methylbutanoic acid | 96309 | 12822 | 64413 | 15171 | NS | 1.50 |
| Dimethyltrisulfide | 7196 | 2755 | 6252 | 3260 | NS | 1.15 |
| **p-Cresol** | **347725** | **27566** | **72516** | **32616** | ** | **4.8** |
| **Indole** | **19943** | **2487** | **6690** | **2942** | (*) | **3.0** |
| **3-methylindole = skatole** | **25322** | **4954** | **3740** | **5862** | ** | **6.8** |

Although the sensory impression of a mixture of odourous compounds is a combination of all compounds in the mixture, some of the compounds can have a higher impact on the odour impression due to their low threshold values. In addition to the threshold values of the odourous compounds the odour quality of the compounds should be taken into consideration. The odour quality of a compound can change by concentration e.g. skatole has a pleasant flower-like odour at very low concentrations whereas the same compound is nauseating at higher concentrations. In contrast some groups of compounds have a relatively pleasant odour description, even at higher concentrations e.g. esters, which usually have

fruity odour notes. By dividing the raw GC-MS data by the odour thresholds of selected compounds we try to illustrate the impact of odours with widely different odour thresholds (Table 2 and Figure 2 and 3). As the reported values in the literature of odour thresholds can vary widely the Figures 2 and 3 is illustrating the extremes. By incorporating the odour thresholds in the raw data an illustration of the impact of sensory impression of the mixture is created, in contrast to the individual compounds. In both figure 2 and 3 the chicory fed pigs are more confined than in the raw data, in contrast to the control fed pigs, which are more scattered. The chicory roots are therefore able to control the production of odorous compounds in the colon, and effectively turn the fermentation from protein fermentation to carbohydrate fermentation.

**Table 2. Odour descriptor and odour thresholds in air of chemical compounds.**

| | Odour descriptor | Odour threshold air mg/m3 Low | Odour threshold air mg/m3 High |
|---|---|---|---|
| Dimethylsulfide (methylthiomethane) | Cooked vegetable, garlic, hydrogen sulfide (1) | 0,002 | 0,65 |
| 2-Butanon | Acetone, varnish (1) | 0,75 | 250 |
| Acetic acid | Vinegar (1) | 0,025 | 76 |
| 2-Pentanon | Jasmine, Geranium, varnish (1) | 11 | 48 |
| Dimethyldisulfide (methyldithiomethane) | Decayed vegetables (3) | 0,003 | 0,029 |
| 1-Pentanol | Alcohol, medicinal (1) | 0,1 | 1100 |
| 2-Methylpropanoic acid (isobutyric acid) | Sweaty, bitter, sour (1) | 0,00072 (3) | 0,0072 (3) |
| Ethylbutyrate (Ethylbutanoate) | Butter, sweetish, apple, perfumed (1) | 0,13 | 0,28 |
| Propylpropionate (propylpropanoate) | Complex fruity odour (apple banana) (2) | 0,23 | 0,26 |
| Butyric acid (butanoic acid) | Buttery, cheesy, sweaty (1) | 0,0004 | 9 |

| | | | |
|---|---|---|---|
| Butanoic acid,2-methyl-,ethylester (*) | | | |
| Propylbutyrat | Pineapple, apricot (2) | | |
| 3-Methylbutanoic acid (isovaleric acid) | Cheese, sweaty (1) | 0,005 | 3 |
| Dimethyltrisulfide (methyltrithiomethane) | Fresh onion (2) | 0,0073 | 0,0073 |
| p-Cresol (4-methyl-phenol) | Phenol like (2) | 0,00005 | 0,04 |
| Indole | Floral (highly pure) otherwise fecal (2) | 0,0006 | 0,0006 |
| 3-Methylindole | Fecal (high concentration) floral (low concentration) (2) | 0,00035 | 0,1 |

Gemert+Nettenbreijer, 1977

(1): Meilgaard, 1975

(2): Fenaroli's Handbook of Flavor Ingredients 3. Ed. 1995

(3): Zahn et al. 2001

(*) Ethyl-2-methylbutyrate is mentioned in Fenaroli's but not with odour descriptor.

In addition to the reduction of the odorous compounds, the feeding with chicory roots may reduce the production of ammonia. The fermentation of inulin in the caecum and colon of pigs results in production of short chain fatty acids. The higher amount of short chain fatty acids reduces the pH. This reduction has a positive influence on the retention of ammonia in the faeces and manure. This results in an improved environment in the stable and in the surroundings (Lenis and Jongbloed, 1999; Sutton et al. 1999). The ammonia emission is further reduced as the bacteria switch from protein-fermentation to carbohydrate fermentation when feeding with chicory roots. Furthermore, as the bacteria grow the nitrogen will be used for production of proteins in the bacterial biomass and is therefore not available for production of ammonia or odorous compounds.

It is not necessary to completely eliminate the presence of odourous compounds in the colon of pigs to reduce the odour impact on ambient air quality. The reduction should only be sufficient to improve the ambient air quality to an acceptable level. The amount of chicory roots necessary for a sufficient reduction of odourous compounds in the colon contents of pigs remains therefore to be determined. If the amount of chicory roots necessary for sufficient reduction can be reduced the method will be more cost effective. In addition to the odour-reducing effects the chicory roots have following benefits: Easy to grow in the present agricultural systems, can be handled by equipment used for other crops as sugar beets, is in it self a valuable feed component, and contain bioactive secondary metabolites (Bais and Ravishankar, 2001).

**Table 3. Experimental design for the final feeding period of the 2 treatments feeding with or without the chicory roots for different periods from 55 –120 kg live weight (9 weeks).**

| Treat-ment | No. of pigs | Food composition and energy level compared to semi ad lib. (100 %) (from 55 – 120 kg) | Roughage |
|---|---|---|---|
| 1 | 8<br>4 female + 4 male | 100 % organic concentrate | None |
| 3 | 8<br>4 female + 4 male | 70 % Organic concentrate + chicory roots (25 %) from 55 kg until slaughter | Chicory roots (2.6-3.0 kg per day) from 55 kg until slaughter |

**Collection of samples and sample preparation**

Immediately after slaughter, the gastrointestinal tract (GIT) was removed and the colon and rectum was separated from the rest of the GIT. The contents from colon and rectum was quantitatively transferred to a basket and mixed so a representative sample could be obtained. The samples were stored at –20°C before preparation for analysis. To prepare the samples for analysis 3 gram were transferred to 10 ml vials

with addition of 3 ml saturated NaCl, the samples were mixed and stored at –80°C before analysis. The saturated NaCl was added to increase the transfer of volatiles to the gas phase and to stop further microbial activity in the samples. On the day of analysis the samples were transferred to an oven hold at 40°C (approximately the body temperature of pigs) and thawed and equilibrated at this temperature for 25 minutes with occasional shaking to increase the transfer of volatiles from the medium to the headspace. For extraction a solid phase microextraction (SPME) fiber (75 $\mu$m polydimethylsiloxane/carboxen, Supelco) was exposed to the headspace for 1 minute and immediately transferred to the injection port of the gas chromatograph for desorption.

**GC-MS measurement of volatiles**

The gas chromatograph was a Varian model STAR 3400 CX. The column was a HP5-MS (Agilent) 30 m long, 0.25 mm internal diameter and with a 0.25 $\mu$m film thickness. Injection temperature was set to 250°C and the column temperature program was as follows: Hold at initial temperature 35°C for 10 minutes, then increase to 130°C with 3°C/minute, finally increase to 250°C with a rate of 40°C/minute and hold at this temperature for 5.34 minutes. The carrier gas was helium with a linear flow rate of 29 cm $s^{-1}$ at 35°C, the samples were run one at a time to secure the samples were treated in exactly the same way. The temperature of the transferline between the gas chromatograph and the mass spectrometer was set to 275°C. The mass spectrometer was a Varian model Saturn 2000 operated in electron impact mode, with the following settings: detection mass range: 35 to 300 m/z; mulitplier voltage: 1800, axial modulation: 4V, trap temperature 200°C; and manifold temperature of 52°C.

Identification

The compounds were identified by comparison with standard spectra from NIST/EPA/NIH or by comparison with spectra from original standards.

**Statistical analysis**

The statistical analyses were carried out with the Statistical Analysis System version 8.2 (SAS Institute, 1999-2001 by SAS Institute Inc., Cary, NC, USA). The GLM procedure was used to calculate the least squares means and standard error of the means for the odour impact compounds from colon. The models included the fixed

effect of diet, sex and animal replicate (litter) as well as interaction between diet and sex (model 1).

$$Y = \mu + a_{diet} + b_{litter} + c_{sex} + ac_{diet*sex} + e_{rror} \qquad \text{(model 1)}$$

Y = dimethylsulfide, 2-butanone, acetic acid, 2-pentanone, dimethyldisulfide, 1-pentanol, 2-methylpropanoic acid, ethylbutyrate, propylpropionate, butyric acid, 3-methylbutanoic acid, propybutyrate, ethyl-2-methyl butanoate ethylester, dimethyltrisulfide, p-cresol, indole, and skatole.

The raw data of the GC-MS areas of the odour compounds as well as values corrected for low and high threshold values were analysed by the GLM- model 1 to investigate the effect of the two diets.

Principal component analysis (PCA) were carried out also using the data of the raw GC-MS area, as well as data corrected for low and high odour threshold, to investigate the effect of the two diets. Full cross validation (leave one out) was applied. Data analysis was carried out with the software The Unscrambler version 7.8 (Camo AS, Oslo, Norway).

**Results**

Table 1 show the peak mean area of GC-MS analyses of selected odour impact compounds found in headspace over the colon samples. The compounds 2-pentanone, ethylbutyrate, propylpropionate, butanoic acid, ethyl-2-methylbutyrate, p-cresol, indole and skatole show significant difference between the two treatments. The esters, which have relatively pleasant odours, are increased in treatment 3 (factorial difference below 1), whereas the malodorous compounds, p-cresol, indole and skatole were decreased in treatment 3 (factorial difference above 1).

The amounts of odour-active compounds found in colon contents does not give a realistic impression of the odour intensity of the mixture as the various compounds can have very different odour thresholds and odour descriptors. Table 2 shows odour threshold values and odour descriptors of the selected compounds found in colon contents. The relative odour activity of the individual compounds can be calculated by dividing the area of the compound with the odour threshold. Thereby can a compound, which is present in low amount result in a high odour impact if the

odour threshold is low. The relative "odour-activity" of the two experimental treatments can therefore be compared. It has not been possible to find odour threshold values for ethyl-2-methylbutyrate and propylbutyrate they are therefore omitted in the calculations.

Figure 1 shows the PCA analysis of the dataset from the raw data. Treatment 1 (control) and treatment 3 (chicory addition) are clearly separated with no overlap between the treatments. The first principal component (x-axis) is controlled by p-cresol (protein degradation product) whereas the second (y-axis) is controlled by butyric acid and propyl propionate which both are degradation products of carbohydrate.

The raw data does not give an impression of the odour of a mixture of volatile compounds, as the compounds can have widely different odour thresholds. The raw data was therefore divided by the odour threshold values found in the literature (Gemert and Nettenbreijer, 1977 and Zahn et al. 2001). The values found in the literature wary widely, the lowest and highest values have therefore both been applied to give an impression of the effect on the potential odour impression. Figure 2 shows the PCA-analysis of the raw data divided by the low odour threshold values to give odour-activity corrected values. The two treatments are clearly separated and the clusters of points are more confined, especially with the pigs given a diet containing chicory. The first principal component is controlled by p-cresol whereas the second is controlled by butyric acid.

Figure 3 shows the PCA-analysis of the raw data divided by the high odour threshold values. The pigs fed control diet are more dispersed and overlap the chicory fed pigs. In contrary to the controls the pigs fed the chicory diet are highly confined. The first principal component is in this case controlled by indole (protein degradation product) whereas the second is controlled by dimethyl disulfide, 2-methyl propanoic acid and to a lesser degree dimethyl trisulfide (all protein degradation products).

**Example 2**

**Influence of chicory roots on boar taint (skatole and androstenone) in pigs**

**Methods**

**Animals and feed**

In 2001, an inulin-rich variety Orchies of chicory (Cichorium intybus L. var. Orchies) for fattening pig diets has been successfully grown at the Research Centre Rugballegård and in 2002 at Research Centre Foulum. The yield of the organically grown crop was 30 t/ha in 2001 and 40 t/ha in 2002. In November 2001, the inulin content (fructan) of the chicory roots of the variety Orchies was around 150 g per kg feed and contained 2.11 MJ net energy (0.27 feed units (FUp)) and 23.4 g digestible protein per kg feed of chicory roots. The pigs ate the high amount of raw and bitter blended chicory roots (from 2.1-3.0 kg per day during the experimental period) without problems after one week of adaptation by giving individually increasing amounts of chicory roots during that week.

The first of two pig experiments began on the 5$^{th}$ November 2001. The experiment consisted of 40 pigs (20 entire male and 20 female pigs), all free of parasite infections. The 40 pigs were kept in litters and fed 100 energy % organic concentrate according to scale (Madsen et al., 1990) and ad libitum grass silage. Composition of the organic concentrate diet during the whole experiment was (g/kg): 145.5 rapeseed cake, 240.0 peas organic, 223.0 wheat organic, 220 barley organic, 50 oat organic, 100.0 GMO-free toasted soybeans, 2 Sv.vit-411 organic, 3.75 salt, 12 limestone and 3.63 monocalcium phosphate. The concentrate diet contained 8.57 MJ net energy (1.11 feed units (FUp)) and 149,7 g digestible protein per kg food. All 40 pigs were then infected with a specific parasite in the period between the 5$^{th}$ November and the 10$^{th}$ December. Eight pigs, four of each sex, were slaughtered on the 10$^{th}$ December due to the parasite experiment.

From the 10$^{th}$ December, the 32 pigs were distributed according to liveweight, litter and sex to four treatments in individual pens (Table 4). Treatment 1 was a conventional control group given 100-energy % organic concentrate and no roughage from the 10$^{th}$ December until slaughter. Treatment 2 was an organic control group given 95 energy % organic concentrate and ad libitum grass silage from the 10$^{th}$ December until slaughter. The 25% blended chicory roots on energy bases plus 70% or-

ganic concentrate were given to treatment 3 from the 10th December until slaughter. However, the first week the pigs had to adapt to eating chicory roots. Treatment 4 was given 95 energy % organic concentrate and semi ad libitum roughage from the 10th December until the 14th January. From the 14th January until the 21st January, the pigs increased the intake of chicory roots (adaptation period), and from the 21st January until slaughter of treatment 4, 25% blended chicory roots on energy bases plus 70% concentrate were given. Blood samples for measuring androstenone and skatole in blood plasma were collected on the 6th and 7th December and the 4th and 6th February of male and female pigs – one week before slaughter. Finally, according to the plan, 16 male pigs were slaughtered on the 11th and 16 female pigs on the 13th February 2002 in order to measure skatole in backfat, and a sensoric panel evaluated eating quality (see Table 4). After one week of adaptation in which the pigs were fed increasing amounts of chicory roots, the pigs ate the high amount of raw and bitter blended chicory roots without problems. The health status and production results of the chicory treatments were as good as the control treatments, and the daily gain corresponded to the results of treatment 2. The chicory-fed pigs ate after the one-week adaptation period 2.1 kg chicory per day from the beginning of treatment 3 and finally 3.0 kg per day during the final three weeks of both treatment 3 and 4. All the planned meat and eating quality measurements have been carried out and analysed. Furthermore, several additional measurements, have been analysed e.g glycogen, driploss, pH, temperature, Minolta colour values L*, a* b* in *M. long. dorsi* and fatty acids.

**Table 4. Experimental design for the final feeding period of the 4 treatments fed diets with or without bioactive chicory roots for different periods from 55-120 kg live weight (9 weeks).**

| Treat-ment | No. of pigs | Food composition and energy level compared with 100 energy % according to scale (55-120 kg) | Bioactive food |
|---|---|---|---|
| 1 Non Bioactive Control | 8<br>4 females +<br>4 males | 100% organic concentrate | None |
| 2<br>Silage | 8<br>4 females +<br>4 males | 95% organic concentrate + ad lib. clover-grass silage | Clover-grass silage from 55 kg until slaughter |
| 3<br>Chicory | 8<br>4 females +<br>4 males | 70% organic concentrate + chicory roots (25%) from 55 kg until slaughter | Chicory roots (2.1-3.0 kg per day) from 55 kg until slaughter |
| 4<br>Chicory/ Silage | 8<br>4 females +<br>4 males | 95% organic concentrate + ad lib. clover-grass silage from 55 kg until 4 weeks before slaughter<br><br>70% organic concentrate + adaptation to chicory roots from 4-3 weeks before slaughter<br><br>70% organic concentrate + chicory roots (25%) the last 3 weeks before slaughter | Clover-grass silage ad lib. from 55 kg until 4 weeks before slaughter<br><br>4-3 weeks before slaughter, adaptation to chicory roots<br><br>chicory roots (25%) (3.0 kg per day) |

**Statistical analysis**

The statistical analyses were carried out with the Statistical Analysis System version 8.2 (SAS Institute, 1999-2001 by SAS Institute Inc., Cary, NC, USA). The GLM procedure was used to calculate the least squares means and standard error of the means for the odour impact compounds from colon. The models included the fixed effect of diet, sex and animal replicate (litter) as well as interaction between diet and sex (model 1).

$$Y = \mu + a_{diet} + b_{litter} + c_{sex} + ac_{diet*sex} + e_{rror} \qquad \text{(model 1)}$$

Y = skatole in blood and backfat and androstenone in blood.

**Results**

The effect of feeding 25% chicory roots plus 70% organic concentrate for a long (treatment 3) or a short time (treatments 4) on skatole and androstenone in blood plasma from *Vena jugularis* and skatole from backfat, and meat and eating quality

have been compared with results of the two control treatments, feeding 100% organic concentrate (treatment 1) or 95% organic concentrate plus clover grass silage (treatment 2) (Table 5 to Table 8).

After one week of adaptation, it was possible to feed 25% minced chicory roots and 70% concentrate on energy bases without problems during the finishing period from 55 kg live weight until slaughter around 120 kg. In the final period, the pigs ate 3 kg minced chicory roots. Some of the pigs found the chicory so palatable that they ate the chicory before the concentrate.

Irrespective of sex and experimental period, all chicory-fed pigs showed skatole concentrations in backfat (after 8 and 3 weeks) and skatole concentrations in blood plasma (after 7 and 2 weeks) which were not significantly different from zero in a statistical GLM analysis in SAS (see Table 5, 6, and 7). A decrease in the androstenone level in treatment 3 compared with treatment 1 seems to be significant, when the results are corrected by the covariate androstenone in blood just before the feeding experiment started. More importantly, none of the chicory-fed male pigs showed androstenone results above the critical limit for off flavour from androstenone as opposed to some of the control-fed male pigs in treatments 1 and 2, which also had skatole concentrations above the off odour limit of 0.20 $\mu$g/g in backfat (see table 8).

**Table 5. Skatole in backfat ($\mu$g/g) according to treatment and sex (Mean and Std. Dev.) (Fjern de ikke signifikante decimaler)**

| Treatment | Sex | N | Mean | Std. dev. |
|---|---|---|---|---|
| 1 | Male | 4 | 0.115 | 0.04358899 |
| 1 | Female | 4 | 0.05 | 0.02828427 |
| 2 | Male | 4 | 0.1325 | 0.11176612 |
| 2 | Female | 4 | 0.0375 | 0.02217356 |
| 3 | Male | 4 | 0.0125 | 0.005 |
| 3 | Female | 4 | 0.0125 | 0.005 |
| 4 | Male | 4 | 0.0175 | 0.00957427 |
| 4 | Female | 4 | 0.01 | 0.00 |

**Table 6. Skatole in backfat (μg/g) (lsmeans and error)**

| Treatment | N | LS Mean | Std. error | Pr > \|t\| |
|---|---|---|---|---|
| 1 | 8 | 0.0825 | 0.01473797 | <.0001 |
| 2 | 8 | 0.085 | 0.01473797 | <.0001 |
| 3 | 8 | 0.0125 | 0.01473797 | 0.4081 |
| 4 | 8 | 0.01375 | 0.01473797 | 0.3639 |

**Table 7. Skatole in blood 1 week before slaughter (μg/l) (lsmens and error)**

| Treatment | N | LS Mean | Std. error | Pr > \|t\| |
|---|---|---|---|---|
| 1 | 8 | 1.8225 | 0.36313839 | 0.0001 |
| 2 | 8 | 2.12875 | 0.36313839 | <.0001 |
| 3 | 8 | 0.0825 | 0.36313839 | 0.8230 |
| 4 | 8 | 0.13 | 0.36313839 | 0.7248 |

**Table 8. Skatole in blood and backfat from the male and female pigs and androstenone in blood from the male pigs plus some performance results in 16 male and 16 female finishing pigs**

| Pig no. | Slaughter date | Treatment* | Feeding[1] | Sex | Live weight | Skatole backfat at slaughter (μg/g) | Skatole blood 1 week before slaughter (μg/l) | Percentage of meat in carcass | Androstenone in blood 1 week before slaughter (ng/ml) |
|---|---|---|---|---|---|---|---|---|---|
| 53 | 11.02.02 | 1 | 100% Concentrate | male | 135.8 | 0.15 | 1.37 | 58.4 | 11.4 |
| 7 | 11.02.02 | 1 | 100% Concentrate | male | 134.5 | 0.15 | 1.72 | 57.2 | 24.4 |
| 27 | 11.02.02 | 1 | 100% Concentrate | male | 112.8 | 0.10 | 1.68 | 59.4 | 11.0 |
| 38 | 11.02.02 | 1 | 100% Concentrate | male | 125 | 0.06 | 0.84 | 58.6 | 19.8 |
| 52 | 13.02.02 | 1 | 100% Concentrate | female | 126 | 0.09 | 2.19 | 57.3 | |
| 9 | 13.02.02 | 1 | 100% Concentrate | female | 123.3 | 0.03 | 2.16 | 59.5 | |
| 34 | 13.02.02 | 1 | 100% Concentrate | female | 113.3 | 0.05 | 3.9 | 60.2 | |
| 50 | 13.02.02 | 1 | 100% Concentrate | female | 116.9 | 0.03 | 0.72 | 61.6 | |
| 54 | 11.02.02 | 2 | 95% Concentrate +silage | male | 113.2 | 0.08 | 1.53 | 59.0 | 7.0 |
| 12 | 11.02.02 | 2 | 95% Concentrate +silage | male | 145.5 | 0.30 | 5.42 | 57.4 | 25.1 |
| 19 | 11.02.02 | 2 | 95% Concentrate +silage | male | 102.5 | 0.08 | 4.44 | 60.0 | 9.6 |
| 36 | 11.02.02 | 2 | 95% Concentrate +silage | male | 120.9 | 0.07 | 0.62 | 60.3 | 9.3 |
| 55 | 13.02.02 | 2 | 95% Concentrate +silage | female | 119.2 | 0.03 | 1.6 | 58.1 | |
| 22 | 13.02.02 | 2 | 95% Concentrate | female | 126.7 | 0.01 | 0.75 | 59.0 | |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | +silage | | | | | | |
| 37 | 13.02.02 | 2 | 95% Concentrate +silage | female | 102.7 | 0.06 | 1.4 | 62.2 | |
| 45 | 13.02.02 | 2 | 95% Concentrate +silage | female | 108.2 | 0.05 | 1.27 | 63.0 | |
| 56 | 11.02.02 | 3 | 70% Conc. + 25% chicory | male | 113 | 0.01 | 0.19 | 59.1 | 6.1 |
| 14 | 11.02.02 | 3 | 70% Conc. + 25% chicory | male | 134 | 0.02 | 0.02 | 57.4 | 16.1 |
| 33 | 11.02.02 | 3 | 70% Conc. + 25% chicory | male | 115.4 | 0.01 | 0.1 | 59.8 | 11.9 |
| 43 | 11.02.02 | 3 | 70% Conc. + 25% chicory | male | 112 | 0.01 | 0.07 | 59.8 | 18.5 |
| 57 | 13.02.02 | 3 | 70% Conc. + 25% chicory | female | 113.8 | 0.01 | 0.05 | 58.7 | |
| 15 | 13.02.02 | 3 | 70% Conc. + 25% chicory | female | 116.5 | 0.01 | 0 | 60.4 | |
| 20 | 13.02.02 | 3 | 70% Conc. + 25% chicory | female | 119.3 | 0.01 | 0.16 | 60.8 | |
| 31 | 13.02.02 | 3 | 70% Conc. + 25% chicory | female | 97.8 | 0.02 | 0.07 | 62.5 | |
| 58 | 11.02.02 | 4 | 70% Conc. + 25% chicory | male | 108.4 | 0.02 | 0 | 60.2 | 10.2 |
| 21 | 11.02.02 | 4 | 70% Conc. + 25% chicory | male | 120.3 | 0.01 | 0.12 | 59.0 | 14.7 |
| 35 | 11.02.02 | 4 | 70% Conc. + 25% chicory | male | 122.4 | 0.03 | 0.24 | 59.1 | 13.7 |
| 49 | 11.02.02 | 4 | 70% Conc. + 25% chicory | male | 116.8 | 0.01 | 0.05 | 60.3 | 10.3 |
| 59 | 13.02.02 | 4 | 70% Conc. + 25% chicory | female | 118.7 | 0.01 | 0.11 | 60.1 | |
| 13 | 13.02.02 | 4 | 70% Conc. + 25% chicory | female | 133.9 | 0.01 | 0.1 | 59.2 | |
| 23 | 13.02.02 | 4 | 70% Conc. + 25% chicory | female | 115 | 0.01 | 0.24 | 60.3 | |
| 51 | 13.02.02 | 4 | 70% Conc. + 25% chicory | female | 96.1 | 0.01 | 0.18 | 62.3 | |

*) Treatment 4 got 25 % chicory roots the last three weeks before slaughter, while Treatment 3 got 25 % chicory roots the last eight weeks before slaughter.

**Example 3**

**The effect of *Cichorium intybus* on helminth infections in pigs**

**Animals and infection**

Five groups of eight parasite naive pigs (four intact males and four females) were infected with 3000 *Oesophagostomum dentatum* L3-larvae while all the animals were on a diet of restricted concentrate + *ad lib* grass silage (Table 9). Four weeks later, one group (infection control group) was slaughtered to assess if the worms had developed to the adult stage and to estimate worm establishment. The animals in the remaining four groups were moved to individual pens and some diets modified. Two groups continued on the concentrate + grass silage diet, while the other two groups were given either concentrate + shredded chicory roots (long term chicory group, 9 weeks) (*Cichorium intybus* L. var. Orchies) or only concentrate (conventional control group). Five weeks later one of the concentrate + grass silage groups had the silage changed to chicory (short term chicory group, 4 weeks), the second group remained on the concentrate + silage diet (organic control group) to the end of the experiment. The surviving pigs were infected a second time with 3000 *O. dentatum* and with 3000 *Ascaris suum* eggs two weeks before slaughter for worm recovery. This was done to examine the effect of diet on both established (= 1st infection, adult worms at slaughter) and establishing (= 2nd infection, immature worms at slaughter) *O. dentatum*. Only the effect on establishing (immature) *A. suum* was investigated in this study .

The pigs used in the experiment were conventionally reared, but all experimental feeds were organically produced. According to the energy level in the feedstuffs 70% and 25% of the daily energy intake was based on concentrate and chicory roots, respectively (Table 9). The total amount of feed given was adjusted according to bodyweight once a week. The pigs fed chicory were adapted to the bitter taste of the root by increasing the chicory proportion to the desired 25% during the first week of the feeding period. At the beginning of the feeding period the long-term chicory group ate 2.1 kg roots and at the end they willingly ate up to 3.0 kg per day.

During the experiment the animals were weighed regularly and faecal samples were collected twice a week for quantification of parasite eggs using a concentration McMaster technique (Nansen & Roepstorff, 1998). Both species of parasite were

recovered from the intestinal contents using an agar-gel technique (Slotved *et al.*, 1996 & 1997).

The data were analysed for an overall difference between groups by the Kruskall Wallis and for differences between individual groups by the Mann Whitney test using the software GraphPad Prism 3.0.

**Results**

At slaughter 4 weeks post infection the infection control group had a mean worm burden (±SD) of 846 ± 748. Of these worms only a mean of 6 worms per pig were not fully mature. The population of adult and immature *O. dentatum* resulting from the first and second experimental infection, respectively, were easily differentiated in all the pigs at the end of the experiment.

Ten days after the introduction of chicory, the long-term chicory group showed a large and rapid reduction in egg excretion compared to the other groups (Figure 4). Though increasing slightly, the egg counts remained at a low level during the remaining part of the experiment. Though a decrease in egg production was also seen in the short-term chicory group, both control groups also showed similar decreases. Overall, the egg excretion converged for all four groups towards the termination of the study. For the first 2½ weeks after the initial diet change the organic control and short term chicory group (both fed concentrate and grass silage in this period) had a higher egg excretion than the conventional control group. Overall, there were unusually large fluctuations in the egg excretion. All eggs were produced by the adult *O. dentatum* that established after the first infection dose, as worms derived from the second infection dose did not fully mature.

At the end of the experiment there was no difference ($p=0.86$) in the populations of established adult *O. dentatum* in the four groups (see Table 10). In contrast, compared to the organic control group, significantly less worms were able to establish in the intestine in both the short ($p=0.04$) and long-term chicory ($p=0.002$) groups. Only the long-term chicory group differed from the conventional control group ($p=0.015$). There was no difference between the conventional and the organic control groups ($p=1.0$). In relation to the organic control group, the population size was reduced with 19% and 33% in the short and long-term chicory groups, respectively. For both

the infection and conventional control group there was an unusually large variation in the establishment of *O. dentatum*.

Overall, there was a statistical difference between the *A. suum* larval counts (Table 10) between the groups (p=0.004). This is primarily due to a significantly smaller recovery of *A. suum* in the long-term chicory group compared to both the conventional (p=0.002) and organic control group (p=0.009). In addition, the short-term chicory group was close to being significantly different from the conventional (p=0.054) and organic control group (p=0.053). No other differences were found. At slaughter four out of the eight long term chicory pigs had a total of 10 adult *A. suum* (5-15 cm) and one pig in the short term chicory group had 1 *A. suum* (3 cm). All 11 worms were older than two weeks and thus not derived from the experimental infections dose.

Generally the pigs had varying degrees of diarrhoea in the early part of the experiment when they were first infected, but production results were satisfactory and identical in all groups, the pigs increasing their mean bodyweight from 55 kg to 120 kg during the experiment. Analysis of the chicory roots showed an inulin content of approximately 150g/kg fresh root.

**Table 9. Diet composition for five groups of pigs. The proportion of feed type is given as % of the daily energy requirement per animal.**

| | Week post first infection | | |
|---|---|---|---|
| Group | -4 – 0 | 0 - 5 | 5 – 9 |
| Infection control | 100% concentrate semi *ad lib* grass silage | - | - |
| "Conventional" control with organic concentrate minus roughage | 100% concentrate semi *ad lib* grass silage | 100% concentrate | 100% concentrate |
| Organic control | 100% concentrate semi *ad lib* grass silage | 95 % concentrate semi ad lib grass silage | 95 % concentrate semi ad lib grass silage |
| Chicory, short term | 100% concentrate semi *ad lib* grass silage | 95 % concentrate semi ad lib grass silage | 70 % concentrate 25 % chicory roots |
| Chicory, long term | 100% concentrate semi *ad lib* grass silage | 70 % concentrate 25 % chicory roots | 70 % concentrate 25 % chicory roots |

**Table 10. Mean worm burden ± SD of *O. dentatum* and *A. suum* in groups of pigs fed different diets. The pigs were infected twice with 3000 *O. dentatum* L3-larvae (11 weeks apart) and once with 2000 *A. suum* eggs. The age of the adult *O. dentatum* populations and the immature *O. dentatum*/*A. suum* populations, are 13 and 2 weeks, respectively.**

| | | *O. dentatum* | | *A. suum* |
|---|---|---|---|---|
| Group | n | Adult | Immature | Immature |
| "Conventional" control with organic concentrate minus roughage | 8 | 1043 ± 975 | 2893 ± 597 | 1072 ± 450 |
| Organic control | 8 | 1281 ± 994 | 3034 ± 479 | 1026 ± 464 |
| Short term chicory | 8 | 989 ± 379 | 2450 ± 469 | 556 ± 302 |
| Long term chicory | 8 | 810 ± 515 | 2017 ± 454 | 288 ± 144 |

**Example 4**

**Sensory profiling of the effects of silage and chicory (bioactive) feeding on boar-taint in cooked pork**

**Sensory characteristics**

Analytical chemists engaged in elucidating boar-taint require clearly defined terminology to describe the sensory characteristics that constitute boar-taint as it is in essence a sensory based off-flavour phenomenon. The development of such descriptors with definitions and references by sensory analysis has much potential in the further elucidation of sensory boar-taint perception and its level of negative effect on consumer acceptability of pork (Bonneau et al., 2000; Dijksterhuis et al., 2000). Sensory profiling, a method by which a panel uses a developed sensory vocabulary to describe perceived sensory characteristics in a sample set has been utilised in the present research (ISO, 1985; ISO, 1994; Meilgaard, et al., 1999; Byrne et al., 2001b). The resultant profile is a perceptual map of the variations in a sample type that can be employed alone or in combination with chemical/instrumental measurements in the explanation and elucidation of underlying sensory and chemical relationships.

The objectives of the present study were to investigate the sensory variation that resulted from the effects of bioactive (silage and chicory) feeding in organically produced male cooked pork. Of particular interest was the effect of bioactive feeding on the 'off-flavour' referred to as boar-taint in the meat. To achieve these aims a descriptive sensory vocabulary was developed with an expert sensory panel and subsequently the panel were utilised to develop a sensory profile for the meat samples, derived from male animals fed various levels of silage and chicory. In the analyses of the sensory profiling data a strategy involving multivariate Principal Component Analysis was utilised to determine precisely how the various feeding treatments were described and discriminated from a sensory perspective.

**Meat preparation**

Pork muscles *Longissimus dorsi* (LD) were used for a sensory analysis. Batches of four muscles each batch from a different animal litter (4 male littermates in each) was obtained. Each of the four muscles in a batch, were from an animal subjected fed one of four treatments prior to slaughter (Table 4 of example 2, although the female were not analysed).

All muscles were stored vacuum packed in darkness at –20°C. Muscles were held at 4°C for approx. 12 h prior to handling to allow ease of cutting and grinding. Visible fat and connective tissues were removed and muscles were cut into cubes (approx. 3 $cm^3$) and mixed thoroughly. Muscles from a specific treatment were utilised, and mixed together thoroughly once cubed. Each treatment batch of muscle cubes was ground in a rotary screw mincer (Model X 70, Scharfen GmbH & Co. Maschinenfabrik KG, Germany) through a 4.5 mm plate. The minced samples were shaped into patties of 100 g and approx. 1 cm thickness using a commercial pattie maker (i.d. 9 cm). Plastic packaging film was used in the making of the patties to help maintain their shape prior to vacuum bagging. Patties were subsequently removed from their plastic film wrapping and vacuum packed in oxygen impermeable plastic laminate bags. The vacuum-packed patties were then frozen at –30°C and stored for up to a week.

Prior to heat treatment, all patties were placed in a 25°C water bath until a core temperature of between 18 and 20°C had been reached. Subsequently patties were removed from their plastic vacuum bags and batch cooked in convection ovens set

to 150°C. The ovens utilised were determined to have comparable heating cycles. The heating/cooking process took a total of 20 min and was carried out as per Byrne et al. (1999b). In each oven, a control patties core temperature was monitored throughout the heat treatment by a termocouple and data logger (Squirrel Series 1000, Grant Instruments Ltd., United Kingdom). The final internal temperature reached over all pattie batches was found to vary between 78 and 82°C. After cooking the samples were cooled to 5°C in oxygen impermeable plastic laminate bags for a short period (10-15 min) prior to reheating for sensory assessment.

To prepare the samples for descriptive vocabulary development and sensory profiling, patties were divided into 8 equal triangular pieces, which were then vacuum packaged in plastic laminate bags. These were placed in a steel tray filled with water at ambient temperature. For reheating the tray was placed in a convection oven at 140°C for 19 min. The mean serving temperature of the vacuum packed samples was 65°C.

**Sensory measurements**

Prior to sensory profiling a sensory panel (8 persons) participated in the development of a sensory vocabulary to describe and discriminate the effects of conventional and bioactive feeding on the general flavour and in particular boar-taint in the pork meat of the present study (see Byrne et al., 1999a,b; Byrne et al., 2001a). The panel was recruited from the public and students of the Royal Veterinary and Agricultural University, Frederiksberg, Denmark. All sensory work was carried out in the sensory laboratory at the University, which fulfils requirements according to the international standards (ASTM, 1986; ISO, 1988).

Panel input, panel leader input, and multivariate statistical analyses were utilised to select a set of 24 descriptors plus an acceptability question from initial list of 32 terms (see Byrne et al., 2001a). Each of the final list of terms was defined by a reference material and terms were divided into their mode of sensory assessment, i.e. odours, tastes, flavours and aftertastes (Table 11).

**Table 11**

**List of 25 sensory descriptive terms with definitions developed for the evaluation of pork meat, oven cooked at 150°C for 20min., derived from male animals fed bioactive compounds, silage and chicory.**

| Term[a b] | Definitions and reference materials[c] |
|---|---|
| **Odour** | **Odour associated with:** |
| 1.Piggy/Animaly-O | cooked pork containing boar-taint/dilute skatole solution |
| 2.Meat/Gamey-O | cooked game meat/wild boar |
| 3.Cardboard -O | wet cardboard |
| 4.Fresh cooked pork meat -O | oven cooked pork meat with on surface browning |
| 5.Linseed oil-O | warmed linseed oil/linseed oil based paint |
| **Taste** | Taste sensation associated with: |
| | sucrose, 1g/l solution in water[c] |
| 7.Sour-T | citric acid (monohydrate) 0.3g/l solution in water |
| 8.Salt-T | sodium chloride, 0.5g/l solution in water |
| 9.Bitter-T | quinine chloride, 0.05g/l solution in water |
| **Flavour** | **Aromatic taste sensation associated with:** |
| 10. Piggy/Animaly-F | cooked pork containing boar-taint/diluted skatole solution |
| 11. Metallic-F | ferrous sulphate, 0.1g/l solution in water |
| 12. Meat/Gamey-F | cooked game meat/wild boar |
| 13. Herby-F | dried mixed herbs |
| 14. Spicy-F | mixed spices |
| 15. Cooked Ham-F | cooked ham |
| 16. Fresh cooked pork meat -F | oven cooked pork meat with no surface browning |
| 17. Cardboard-F | wet cardboard |
| 18. Lactic/Fresh sour -F | natural yoghurt |
| 19. White pepper-F | white pepper |
| 20. Pork fat-F | cooked pork fat |
| **Aftertaste** | **Feeling factor in the oral cavity associated with:** |
| 21. Lactic/Fresh sour -AT | natural yoghurt |
| 22. Astringent-AT | aluminium sulphate 0.02g/l solution in water |
| 23. Spicy/heat-AT | mild warming effect of spices |
| 24. Chemical medicinal | cough syrup |
| 25. Fatty mouth coating-AT | a residual coating of fat after sample assessment |
| 26. Acceptibility | how acceptable do you find the sample? |

[a] Suffix to sensory terms indicates method of assessment by panellists; -O = Odour, -F = Flavour,-T = Taste, -AT = Aftertaste.

[b] Concentrations in g/l were devised to ensure panellists' could recognise clearly the sensory note involved.

[c] Definitions of sensory terms as derived during vocabulary development.

A sensory profile was developed for the pork patties for each of the 4 feeding treatments using the same 8-member paid panel as utilised in vocabulary development. The sample set presented at the profiling study contained the four treatments. This sample set (4) was assessed by each of the 8 panel assessors 4 times, as replicates (4 × 8 × 4) = 128 'objects' in the profile data set for each of the 25 sensory descriptors. Each replicate was presented on each of 4 days to each panellist, 4 samples per day. In all 4 days of panel sessions of 1.5 hr each were carried out in the development of the profile. Presentation to individual panellists on each day of profiling was in a randomised order. However, the full range of storage days and feeding treatments was included on each day.

Quantitative data was collected using the FIZZ Network data acquisition software (BIOSYSTEMS, Couternon, France). Unstructured line scales of 15 cm anchored on the left side by the term 'none' and on the right side by the term 'extreme' were used for the scoring of each sensory term (Meilgaard et al., 1999).

All multivariate analyses were performed using the Unscrambler Software, Version 7.5 (CAMO ASA, Trondheim, Norway). In PCA analysis, data were analysed, centred with full cross-validation.

**Results**

Multivariate Principal Component Analysis was used to gain a qualitative overview of the relationships within the sensory data and the association of the descriptors with the experimental design variables, i.e. non-bioactive/control, silage, chicory/silage and chicory feeding.

A sensory profiling of cooked pork derived from male animals was illustrated by Principal Component Analysis (PCA) plot (Figure 5). PCA was found present 2 significant Principal Components (PCs). PC1 and PC2 explaining 43 and 33% of the explained variation, respectively.

The general sensory description of the feeding treatments is shown in Table 5 of example 2 and Figure 5.

1. Non-bioactive control diet (100% organic concentrates):
These samples were described by pork meatiness-flavour, sweet-taste, pork fat-flavour, salt-taste. These are typical 'fresh' and sweet meaty attributes of conventional feeding (Byrne et al., 2001b). However, associated with this aspect of the samples was a high level of boar-taint as described by Piggy/Animaly-flavour and odour.

2. Control + silage:
These samples appear not as 'fresh' in relation to meatiness as control diet and contain a number off-flavours, i.e. cardboard odour/flavour and linseed oil-like odour.

3. and 4. Control + chicory and control +silage +chicory, respectively:
These samples are described as having pork meatiness-odour, meat/gamey-flavour, spicy-aftertaste, herby-flavour, sour-taste, bitter-taste, astringent-aftertaste. These diets appear to have no off-flavours and have 'fresh' meat character as per the non-bioactive diet. Also 'pleasent' herby and spicy characteristics are present.

Overall, feed 3. Control + chicory was perceived as the most acceptable in its sensory characteristics relative to the other feeding treatments

Chicory and Silage/Chicory are similar in their sensory characteristics (bitter tasting and have freshly cooked meat odour), and are negatively correlated to boar-taint as described by Piggy/Animaly-flavour and odour. Thus, the chicory treatments are more acceptable as they have reduced boar-taint from a sensory perspective (Figure 5).

The non-bioactive control feeding treatment is the most boar tainted as indicated by the samples positive correlation to the descriptors Piggy/Animaly-flavour and odour.

Control and Silage have many common sensory characteristics, however, Silage appears to be related somewhat to have more lipid oxidation based off-note descriptors (cardboard and linseed-oil like), even in freshly cooked samples as were the samples in the present study. This was most likely related to higher levels of unsaturated phospholipids in the meat elevated through silage feeding. Thus, the

silage fed samples had an increased potential for lipid-oxidation relative to all other feeding treatments (Byrne et al., 2001b).

**Conclusions**

Treatments 3. chicory and 4. silage/chicory are very similar and are much lower in boar-taint from a sensory perspective than treatments 1. Non-bioactive and 2. Silage. Treatment 2. silage also appears to be the most lipid-oxidative of the samples.

Overall, chicory appears to reduce boar-taint and this clearly noted by the sensory panel.

The most important aspect of this is the panel has indicated that the chicory effect on reducing boar-taint results in acceptable fresh pork meat from a sensory perspective.

The main point of course being that chicory having clearly reduced boar-taint from a sensory perception perspective did not lead to the imparting of other off-flavours in the freshly cooked meat of the chicory fed samples.

The non-bioactive control fed pigs were found to have a higher level of boar-taint as described by the term Piggy/Animaly-odour and flavour relative to the pigs fed chicory. Thus, the chicory fed pigs had a more acceptable sensory character than the pigs feed non-bioactive control from a 'consumer' perspective, in relation to boar-taint.

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## Claims

1. A method for reducing taint in animals, said method comprising feeding to a male animal a chicory root product during at least two days prior to slaughtering the animal.

2. The method of claim 1, wherein the chicory root product is fed to the animal for at least one week, for example at least 1.5 weeks, such as at least 2 weeks, preferably at least 3 weeks, such as at least 4 weeks, for example at least 5 weeks, such as at least 6 weeks, for example at least 7 weeks, such as at least 8 weeks, for example at least 9 weeks, such as at least 10 weeks, for example at least 15 weeks, such as at least 20 weeks.

3. The method of claim 1-2, wherein the chicory root product is fed to the animal substantially until slaughter.

4. The method according to any of the preceding claims, wherein the chicory root product is fed to the animal daily.

5. The method of claim 4, wherein the chicory root product is fed to the animal several times daily, such as 2 times, 3 times, 4 times, 5 times, or more than 5 times.

6. The method according to any of the preceding claims, wherein the chicory root product part of the ration of the animal is at least 5 % on a daily energy basis.

7. The method of claim 6, wherein the chicory root product part of the ration of the animal is at least 10 % on a daily energy basis.

8. The method of claim 6, wherein the chicory root part comprises at least 15 % of the ration, more preferably at least 20%, more preferably at least 25%, more preferably at least 30 %, for example at least 35%, such as at least 40%, for example at least 50%, such as at least 60%, for example at least 75%, such as at least 90%, for example substantially 100%.

9. The method according to any of the preceding claims, wherein the animal is a ruminant, such as cow, sheep, goat, buffalo.

10. The method according to any of the preceding claims 1 to 8, wherein the animal is a monogastric species, such as horse, pig, poultry, dog, and cat.

11. The method according to claim 10, wherein the monogastric species is a pig.

12. The method according to claim 11, wherein the pig is a male pig.

13. The method according to claim 12, wherein the pig is an entire male pig.

14. The method according to claim 11-13, wherein weight of the pig is from 25 to 300 kg, preferably as from 55 to 160 kg.

15. The method according to any of the preceding claims, wherein the species of Chicory is Cichorium intybus L.

16. The method according to any of the preceding claims, wherein the chicory roots contain at least 5% inulin, more preferably at least 10% inulin, more preferably at least 15 % inulin, more preferably at least 20 % inulin, such as at least 25% inulin, for example at least 30 % inulin.

17. The method according to any of the preceding claims, wherein the chicory roots contain at least 5% FOS, more preferably at least 10% FOS, more preferably at least 15 % FOS, more preferably at least 20 % FOS, such as at least 25% FOS, for example at least 30 % FOS.

18. The method according to any of the preceding claims, wherein the chicory root product comprises a silage product of chicory roots, such as a silage product of essentially whole chicory roots.

19. The method according to any of the preceding claims, wherein the chicory root product comprises a fermented product of chicory roots.

20. The method according to any of the preceding claims, wherein the chicory root product comprises a dried product of chicory roots, such as a dried product of essentially whole chicory roots.

21. The method according to any of the preceding claims, wherein the chicory root product is a disintegrated product, such as a powder, flakes, pulp, slices, flour, pellets.

22. The method according to any of the preceding claims, wherein the chicory root product comprises fresh chicory roots.

23. The method according to any of the preceding claims, wherein the chicory root product comprises a fraction and/or an extract of chicory roots.

24. The method according to claim 23, wherein the fraction and/or extract comprises an inulin fraction and a low molecular weight fraction comprising coumarins and/or sesquiterpenes.

25. A method for reducing the skatole content in animals, said method comprising feeding to a animal a chicory root product for at least two days prior to slaughtering.

26. The method of claim 25, wherein the skatole content of blood is reduced by at least 25%, more preferably at least 40%, more preferably at least 50%, more preferably at least 75%, more preferably at least 80%, more preferably at least 90%, more preferably at least 95%, more preferably at least 98%, more preferably to substantially 0.

27. The method of claim 25, wherein the skatole content of blood and/or backfat is reduced to below the human sensory threshold.

28. The method of claim 25, wherein the skatole content of backfat is reduced by at least 25%, more preferably at least 40%, more preferably at least 50%, more preferably at least 75%, more preferably at least 80%, more preferably at least

90%, more preferably at least 95%, more preferably at least 98%, more preferably to substantially 0.

29. The method of claim 25-28, further including the features of claim 2 to 24.

30. A method for reducing the androstenone content in meat and/or fat and/or blood said method comprising feeding to an animal a chicory root product for at least two days.

31. The method of claim 30, wherein the androstenone content is reduced by at least 10%, more preferably at least 25%, more preferably at least 40%, more preferably at least 50%, more preferably at least 75%, more preferably at least 80%, more preferably at least 90%, more preferably at least 95%, more preferably at least 98%.

32. The method of claim 30-31, wherein the androstenone content in meat and/or fat is reduced to below the human sensory threshold.

33. The method of claim 30-32, wherein the animal is subsequently slaughtered.

34. The method of claim 30-33, further including the features of claim 2 to 24.

35. A method for improving the sensory characteristics comprising odour, flavour, taste and/or aftertaste of meat from a human sensory perspective, said method comprising feeding to an animal a chicory root product for at least two days prior to slaughter.

36. The method of claim 35, wherein the improvement of sensory characteristics is a reduction of boar taint comprising reducing Piggy/Animaly-odour and/or Piggy/Animaly-flavour to an acceptable level from a human sensory perspective.

37. The method of claim 35, wherein the improvement of sensory characteristics is a reduction of boar taint comprising increasing acceptable sensory characteristics selected from the group of sour-taste, bitter-taste, pork meatiness-odour, astringent-taste, metallic-flavour and Fresh sour/lactic-flavour.

38. The method of claim 35, wherein the improvement of sensory characteristics comprises reduction of sensory characteristics selected from the group of: Piggy/Animaly-odour, Meat/Gamey-odour, Cardboard-like-odour, Linseed oil-odour, Sweet-taste, Salt-taste, Piggy/Animaly-flavour, Cooked ham-flavour, Fresh cooked pork meat like-flavour, Cardboard-flavour, White pepper-flavour, Pork fat-flavour, Lactic/Fresh sour-aftertaste, spicy heat aftertaste, Chemical/medicinal-aftertaste, Fatty mouth coating-aftertaste, Unacceptability.

39. The method of claim 35, wherein the improvement of sensory characteristics comprises increasing of sensory characteristics selected from the group of: Fresh cooked pork meat like-odour, Sour-taste, Bitter-taste, Metallic-flavour, Meat/Gamey-flavour, Herby-flavour, Spicy-flavour, Lactic/fresh sour-flavour, Astringent-aftertaste, Acceptability.

40. The method of claim 35-39, further including the features of claim 2 to 24.

41. A method for reducing malodour in the environment, said method comprising feeding a chicory root product to animals for at least two days.

42. The method according to claim 41, wherein the reduction is caused by a relative reduction in skatole and/or p-cresole and/or indole in the gastrointestinal tract.

43. The method according to claim 41-42, wherein the reduction is caused by a relative increase in the amount of 2-pentanon and/or ethylbutyrate and/or propylpropionate and/or propylbutyrate and/or butanoic acid 2-methyl-ethylester in the gastrointestinal tract.

44. The method according to claim 41-43, wherein the animal is a ruminant such as cattle, buffalo, sheep, goat.

45. The method according to claim 41-43, wherein the animal is a monogastric species.

46. The method of claim 45, wherein the monogastric animal is a furred animal, such as mink, fox, muskrat, rabbit, hare, wolf.

47. The method of claim 45, wherein the monogastric animal is an animal used for meat, such as pig, poultry, rabbit, hare, more preferably wherein the monogastric animal is a pig.

48. The method according to any of the preceding claims 41 to 47, wherein the malodour is stable malodour and the animal is kept in a stable.

49. The method according to claim 48, wherein the animal is kept in the stable for at least 8 hours a day.

50. The method according to any of the preceding claims 41 to 49, wherein the malodour is manure malodour and the manure originates from animals fed with the chicory root product.

51. The method of claim 41 to 50, further including the features of claim 2 to 24.

52. A method for reducing the amount of infections of the gastrointestinal tract in a non-human animal, said method comprising feeding to a non-human animal a chicory root product for at least two days.

53. The method of claim 52, wherein the infections are parasites.

54. The method of claim 53, wherein the parasites are worms.

55. The method of claim 53, wherein the reduction is a reduction of the number of eggs in the animal faeces.

56. The method of claim 52, wherein the infections are microbiological infections selected from Coli, Salmonella, Campylobacter and Yersinia

57. The method of claim 54, wherein the infections are worms selected from *Ascaris suum*, *Oesophagostomum dentatum*, *Oesophagostomum quadrispinulatum*,

*Oesophagostomum brevicaudum*, *Oesophagostomum granatensis*, *Oesophagostomum georgianum*, *Hyostrongylus rubidus*, *Trichuris suis*, and *Strongyloides ransomi* and *Trichinella* spp.

58. The method of claim 52 to 57, further including the features of claim 2 to 24.

59. Use of chicory roots as a feed product for "grown up" (>> 7 weeks) pigs.

60. Use of chicory roots for preparing a feed product for "grown up" pigs.

61. Use of chicory roots for preparing a product for the prevention of boar taint.

62. Use of chicory roots for preparing a product for reduction of skatole content in pigs, in particular in boar fat.

63. Use of chicory roots for preparing a product for reduction of androstenone in pigs.

64. Use of chicory roots for preparing a product for reduction or prevention of gastrointestinal tract infections in pigs.

65. The use according to claim 59-64 further including the features of claim 2-24.

66. Use of a method for reducing taint in animals, said method comprising feeding to a male animal a chicory root product during at least two days prior to slaughtering the animal.

67. The use of the method of claim 65, further including the features of claim 2-55.

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**Figure 1**

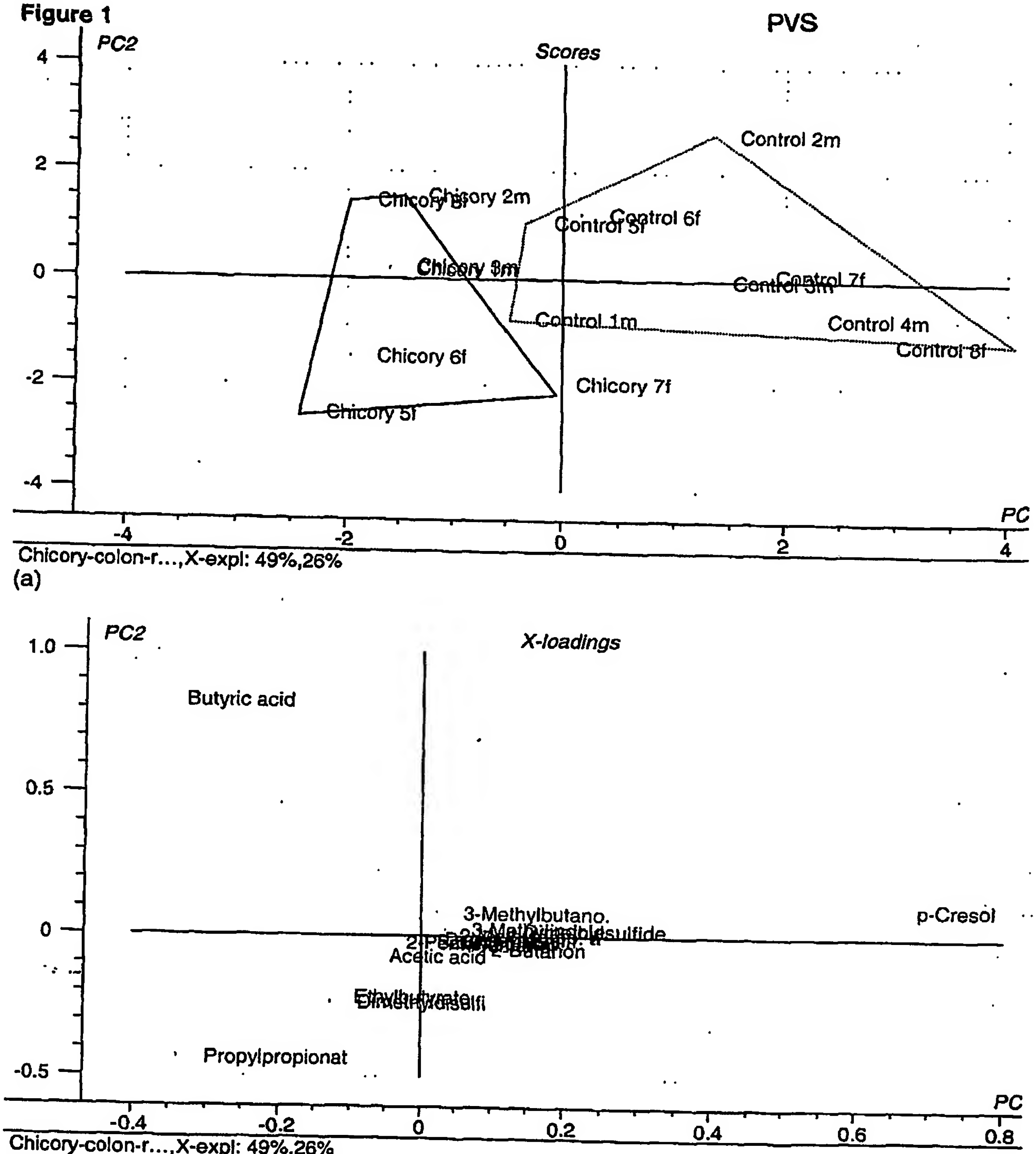


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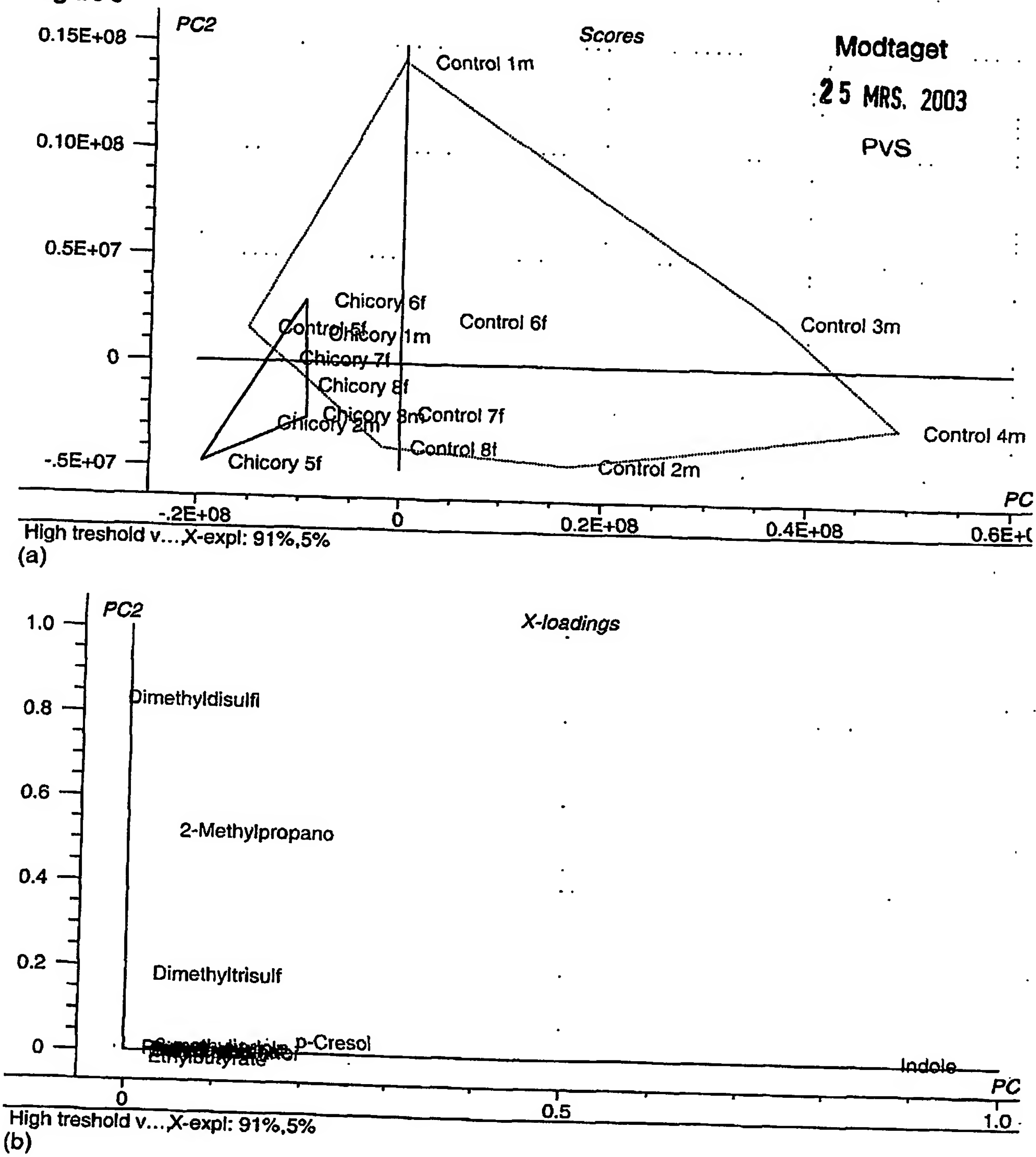
Figure 3

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# Figure 4

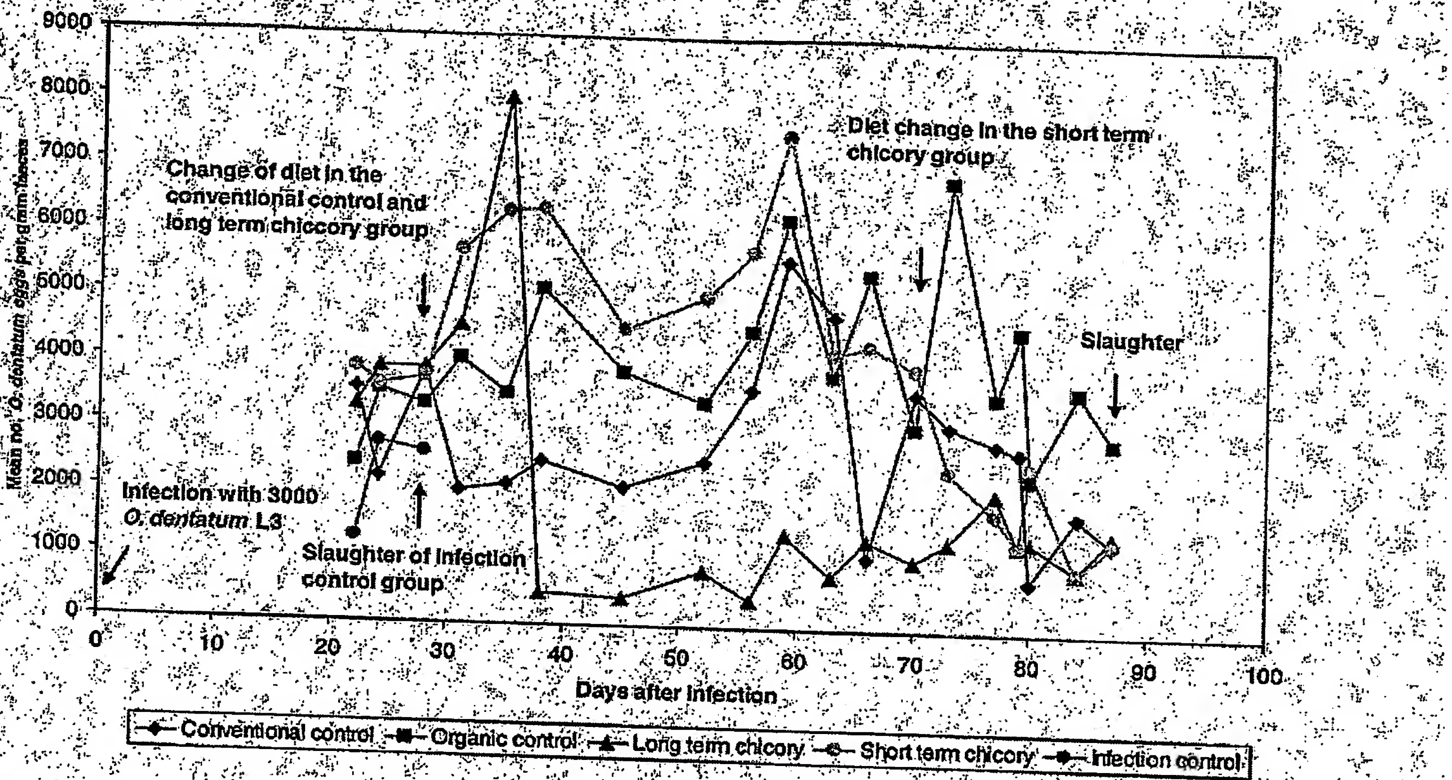

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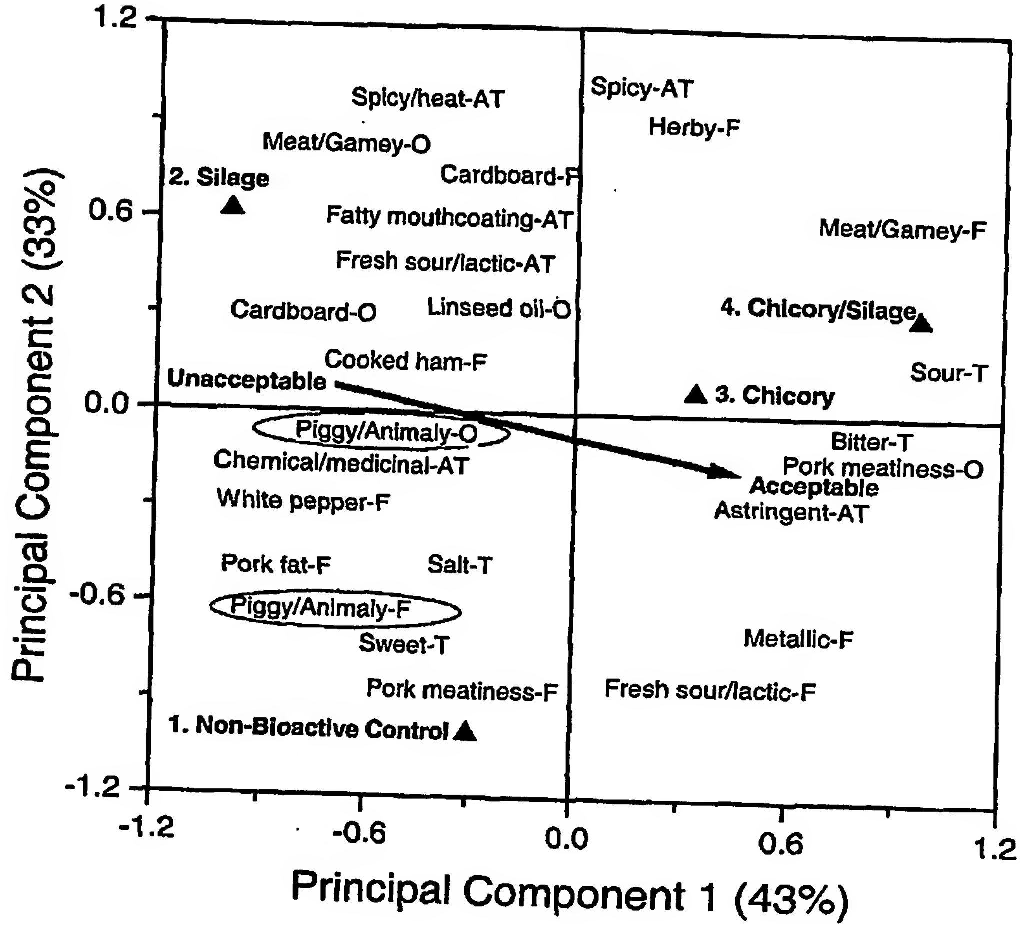
**Figure 5**




Principal Component Analysis (PCA) of sensory profiling data from freshly cooked male pork meat samples for each of four feeding treatments, 1). Non-Bioactive Control, 2). Silage, 3). Chicory, and 4). Chicory/Silage (see Table 5, example 2)

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